```
  * * * * * * * * * * * * * STN  Columbus  * * * * * * * * * * * * * *

FILE 'HOME' ENTERED AT 14:34:34 ON 12 JUL 2004

=> fil .bec
COST IN U.S. DOLLARS                                    SINCE FILE      TOTAL
                                                           ENTRY    SESSION
FULL ESTIMATED COST                                         0.21       0.21

FILES 'MEDLINE, SCISEARCH, LIFESCI, BIOTECHDS, BIOSIS, EMBASE, HCAPLUS, NTIS,
       ESBIOBASE, BIOTECHNO, WPIDS' ENTERED AT 14:34:49 ON 12 JUL 2004
ALL COPYRIGHTS AND RESTRICTIONS APPLY.  SEE HELP USAGETERMS FOR DETAILS.

11 FILES IN THE FILE LIST

=> s vitro(8a)sialyl?
FILE 'MEDLINE'
        767228 VITRO
          6681 SIALYL?
L1          94 VITRO(8A)SIALYL?

FILE 'SCISEARCH'
        388474 VITRO
          6881 SIALYL?
L2          76 VITRO(8A)SIALYL?

FILE 'LIFESCI'
        180174 VITRO
          1641 SIALYL?
L3          26 VITRO(8A)SIALYL?

FILE 'BIOTECHDS'
         22926 VITRO
           424 SIALYL?
L4            5 VITRO(8A)SIALYL?

FILE 'BIOSIS'
        613570 VITRO
          7444 SIALYL?
L5          105 VITRO(8A)SIALYL?

FILE 'EMBASE'
        917533 VITRO
          6329 SIALYL?
L6          98 VITRO(8A)SIALYL?

FILE 'HCAPLUS'
        566820 VITRO
          8296 SIALYL?
L7          124 VITRO(8A)SIALYL?

FILE 'NTIS'
          8635 VITRO
            18 SIALYL?
L8            0 VITRO(8A)SIALYL?

FILE 'ESBIOBASE'
        174930 VITRO
          2801 SIALYL?
L9          47 VITRO(8A)SIALYL?

FILE 'BIOTECHNO'
        253158 VITRO
          3202 SIALYL?
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L10          58 VITRO(8A)SIALYL?

FILE 'WPIDS'
         20819 VITRO
           433 SIALYL?
L11            5 VITRO(8A)SIALYL?

TOTAL FOR ALL FILES
L12          638 VITRO(8A) SIALYL?

=> s l12 not 1998-2004/py
FILE 'MEDLINE'
       3290180 1998-2004/PY
L13           65 L1 NOT 1998-2004/PY

FILE 'SCISEARCH'
       6419509 1998-2004/PY
L14           41 L2 NOT 1998-2004/PY

FILE 'LIFESCI'
        673560 1998-2004/PY
L15           16 L3 NOT 1998-2004/PY

FILE 'BIOTECHDS'
        116634 1998-2004/PY
L16            4 L4 NOT 1998-2004/PY

FILE 'BIOSIS'
       3525824 1998-2004/PY
L17           70 L5 NOT 1998-2004/PY

FILE 'EMBASE'
       2922077 1998-2004/PY
L18           66 L6 NOT 1998-2004/PY

FILE 'HCAPLUS'
       6030194 1998-2004/PY
L19           79 L7 NOT 1998-2004/PY

FILE 'NTIS'
        126221 1998-2004/PY
L20            0 L8 NOT 1998-2004/PY

FILE 'ESBIOBASE'
       1861105 1998-2004/PY
L21           19 L9 NOT 1998-2004/PY

FILE 'BIOTECHNO'
        724097 1998-2004/PY
L22           36 L10 NOT 1998-2004/PY

FILE 'WPIDS'
       5230767 1998-2004/PY
L23            0 L11 NOT 1998-2004/PY

TOTAL FOR ALL FILES
L24          396 L12 NOT 1998-2004/PY

=> s (commercial or scale or batch)(10a)(sialyl? or glycosylat?)
FILE 'MEDLINE'
         41073 COMMERCIAL
        128227 SCALE
         10097 BATCH
          6681 SIALYL?
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         41363 GLYCOSYLAT?
L25         58 (COMMERCIAL OR SCALE OR BATCH)(10A)(SIALYL? OR GLYCOSYLAT?)

FILE 'SCISEARCH'
         92354 COMMERCIAL
        295201 SCALE
         35602 BATCH
          6881 SIALYL?
         32182 GLYCOSYLAT?
L26         93 (COMMERCIAL OR SCALE OR BATCH)(10A)(SIALYL? OR GLYCOSYLAT?)

FILE 'LIFESCI'
         22003 COMMERCIAL
         33693 SCALE
         10590 BATCH
          1641 SIALYL?
          9824 GLYCOSYLAT?
L27         27 (COMMERCIAL OR SCALE OR BATCH)(10A)(SIALYL? OR GLYCOSYLAT?)

FILE 'BIOTECHDS'
          5919 COMMERCIAL
         14743 SCALE
         12379 BATCH
           424 SIALYL?
          3713 GLYCOSYLAT?
L28         66 (COMMERCIAL OR SCALE OR BATCH)(10A)(SIALYL? OR GLYCOSYLAT?)

FILE 'BIOSIS'
         90418 COMMERCIAL
        145421 SCALE
         23754 BATCH
          7444 SIALYL?
         35871 GLYCOSYLAT?
L29         74 (COMMERCIAL OR SCALE OR BATCH)(10A)(SIALYL? OR GLYCOSYLAT?)

FILE 'EMBASE'
         38512 COMMERCIAL
        140556 SCALE
         15930 BATCH
          6329 SIALYL?
         32978 GLYCOSYLAT?
L30         78 (COMMERCIAL OR SCALE OR BATCH)(10A)(SIALYL? OR GLYCOSYLAT?)

FILE 'HCAPLUS'
         28843 COMMERCIAL
        273581 COM
        287330 COMMERCIAL
                 (COMMERCIAL OR COM)
        326677 SCALE
         80903 BATCH
          8296 SIALYL?
         39167 GLYCOSYLAT?
L31        151 (COMMERCIAL OR SCALE OR BATCH)(10A)(SIALYL? OR GLYCOSYLAT?)

FILE 'NTIS'
         52378 COMMERCIAL
         81529 SCALE
          6328 BATCH
            18 SIALYL?
           121 GLYCOSYLAT?
L32          1 (COMMERCIAL OR SCALE OR BATCH)(10A)(SIALYL? OR GLYCOSYLAT?)

FILE 'ESBIOBASE'
         21728 COMMERCIAL
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         53950 SCALE
         10738 BATCH
          2801 SIALYL?
         12537 GLYCOSYLAT?
L33         45 (COMMERCIAL OR SCALE OR BATCH)(10A)(SIALYL? OR GLYCOSYLAT?)

FILE 'BIOTECHNO'
         14938 COMMERCIAL
         23003 SCALE
         11409 BATCH
          3202 SIALYL?
         16990 GLYCOSYLAT?
L34         46 (COMMERCIAL OR SCALE OR BATCH)(10A)(SIALYL? OR GLYCOSYLAT?)

FILE 'WPIDS'
         42017 COMMERCIAL
        122488 SCALE
         27024 BATCH
           433 SIALYL?
          2629 GLYCOSYLAT?
L35         12 (COMMERCIAL OR SCALE OR BATCH)(10A)(SIALYL? OR GLYCOSYLAT?)

TOTAL FOR ALL FILES
L36        651 (COMMERCIAL OR SCALE OR BATCH)(10A)(SIALYL? OR GLYCOSYLAT?)

=> s l36 not 1998-2004/py
FILE 'MEDLINE'
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L37         29 L25 NOT 1998-2004/PY

FILE 'SCISEARCH'
       6419509 1998-2004/PY
L38         45 L26 NOT 1998-2004/PY

FILE 'LIFESCI'
        673560 1998-2004/PY
L39         15 L27 NOT 1998-2004/PY

FILE 'BIOTECHDS'
        116634 1998-2004/PY
L40         47 L28 NOT 1998-2004/PY

FILE 'BIOSIS'
       3525824 1998-2004/PY
L41         42 L29 NOT 1998-2004/PY

FILE 'EMBASE'
       2922077 1998-2004/PY
L42         44 L30 NOT 1998-2004/PY

FILE 'HCAPLUS'
       6030194 1998-2004/PY
L43         74 L31 NOT 1998-2004/PY

FILE 'NTIS'
        126221 1998-2004/PY
L44          1 L32 NOT 1998-2004/PY

FILE 'ESBIOBASE'
       1861105 1998-2004/PY
L45         12 L33 NOT 1998-2004/PY

FILE 'BIOTECHNO'
        724097 1998-2004/PY
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L46          24 L34 NOT 1998-2004/PY

FILE 'WPIDS'
       5230767 1998-2004/PY
L47           3 L35 NOT 1998-2004/PY

TOTAL FOR ALL FILES
L48         336 L36 NOT 1998-2004/PY

=> fil .becpat
COST IN U.S. DOLLARS                                  SINCE FILE      TOTAL
                                                         ENTRY    SESSION
FULL ESTIMATED COST                                      24.18      24.39

FILES 'BIOTECHDS, HCAPLUS, WPIDS' ENTERED AT 14:38:43 ON 12 JUL 2004
ALL COPYRIGHTS AND RESTRICTIONS APPLY.  SEE HELP USAGETERMS FOR DETAILS.

3 FILES IN THE FILE LIST

=> s l12 and wo/pc and pry<=1998 and py>2000 range=2001,
FILE 'BIOTECHDS'
          25454 WO/PC
           1921 PRY<=1998
                  (PRY<=1998)
          73863 PY>2000
                  (PY>2000)
L49           0 L4 AND WO/PC AND PRY<=1998 AND PY>2000

FILE 'HCAPLUS'
         174812 WO/PC
          41174 PRY<=1998
        3286603 PY>2000
L50           0 L7 AND WO/PC AND PRY<=1998 AND PY>2000

FILE 'WPIDS'
         376350 WO/PC
         194245 PRY<=1998
                  (PRY<=1998)
        2607983 PY>2000
                  (PY>2000)
L51           0 L11 AND WO/PC AND PRY<=1998 AND PY>2000

TOTAL FOR ALL FILES
L52           0 L12 AND WO/PC AND PRY<=1998 AND PY>2000

=> s l36 and wo/pc and pry<=1997 and py>2000 range=2000,
FILE 'BIOTECHDS'
          30362 WO/PC
           2094 PRY<=1997
                  (PRY<=1997)
          73876 PY>2000
                  (PY>2000)
L53           0 L28 AND WO/PC AND PRY<=1997 AND PY>2000

FILE 'HCAPLUS'
         210239 WO/PC
          51324 PRY<=1997
        3354474 PY>2000
L54           0 L31 AND WO/PC AND PRY<=1997 AND PY>2000

FILE 'WPIDS'
         451438 WO/PC
         239680 PRY<=1997
                  (PRY<=1997)
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```
       2769117 PY>2000
                 (PY>2000)
L55            0 L35 AND WO/PC AND PRY<=1997 AND PY>2000

TOTAL FOR ALL FILES
L56            0 L36 AND WO/PC AND PRY<=1997 AND PY>2000

=> log y
COST IN U.S. DOLLARS                          SINCE FILE      TOTAL
                                                 ENTRY    SESSION
FULL ESTIMATED COST                              15.23      39.62

STN INTERNATIONAL LOGOFF AT 14:40:50 ON 12 JUL 2004
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| Ref # | Hits | Search Query | DBs | Default Operator | Plurals | Time Stamp |
|---|---|---|---|---|---|---|
| L1 | 32 | vitro near4 sialyla$ | US-PGPUB; USPAT | OR | OFF | 2004/07/12 14:11 |
| L2 | 38 | (commercial or scale or batch) near4 sialyl$ | US-PGPUB; USPAT | OR | OFF | 2004/07/12 14:20 |

1/15/98

| Ref # | Hits | Search Query | DBs | Default Operator | Plurals | Time Stamp |
|---|---|---|---|---|---|---|
| L1 | 32 | vitro near4 sialyla$ | US-PGPUB; USPAT | OR | OFF | 2004/07/12 14:11 |

PGPUB-DOCUMENT-NUMBER: 20040132640

PGPUB-FILING-TYPE: new

DOCUMENT-IDENTIFIER: US 20040132640 A1

TITLE: Glycopegylation methods and proteins/peptides produced by the methods

PUBLICATION-DATE: July 8, 2004

INVENTOR-INFORMATION:

| NAME | CITY | STATE | COUNTRY | RULE-47 |
|---|---|---|---|---|
| DeFrees, Shawn | North Wales | PA | US | |
| Zopf, David | Wayne | PA | US | |
| Bayer, Robert | San Diego | CA | US | |
| Bowe, Caryn | Doylestown | PA | US | |
| Hakes, David | Willow Grove | PA | US | |
| Chen, Xi | Lansdale | PA | US | |

APPL-NO: 10/ 411012

DATE FILED: April 9, 2003

RELATED-US-APPL-DATA:

child 10411012 A1 20030409

parent continuation-in-part-of PCT/US02/32263 20021009 US PENDING

non-provisional-of-provisional 60407527 20020828 US

non-provisional-of-provisional 60404249 20020816 US

non-provisional-of-provisional 60396594 20020717 US

non-provisional-of-provisional 60391777 20020625 US

non-provisional-of-provisional 60387292 20020607 US

US-CL-CURRENT: 514/8, 530/395

ABSTRACT:

The invention includes methods and compositions for remodeling a peptide molecule, including the addition or deletion of one or more glycosyl groups to a peptide, and/or the addition of a modifying group to a peptide.

CROSS REFERENCE TO RELATED APPLICATIONS

[0001] This application is a continuation-in-part of prior Application No. PCT/US02/32263, filed Oct. 9, 2002; Provisional Patent Application No. 60/448,381, filed Feb. 19, 2003 (converted to non-provisional application, same filing date, serial number not yet assigned); Provisional Patent Application No. 60/438,582, filed Jan. 6, 2003 (converted to non-provisional application, same filing date, serial number not yet assigned); Provisional Patent Application No. 60/407,527, filed Aug. 28, 2002; Provisional Patent Application No. 60/404,249, filed Aug. 16, 2002; Provisional Patent

Application No. 60/396,594, filed Jul. 17, 2002; Provisional Patent Application No. 60/391,777, filed Jun. 25, 2002; Provisional Patent Application No. 60/387,292, filed Jun. 7, 2002; Provisional Patent Application No. 60/334,301, filed Nov. 28, 2001; Provisional Patent Application No. 60/334,233, filed Nov. 28, 2001; Provisional Patent Application No. 60/344,692, filed Oct. 19, 2001; and Provisional Patent Application No. 60/328,523, filed Oct. 10, 2001.

---------- KWIC ---------

Detail Description Paragraph - DETX (1059):

[1372] In vitro sialylation was carried out using the G2 glycoform antibody (1 mg/mL) by contacting it with 0.1 U/mg ST3Gal3 or 0.1 U/mg ST6Gal1, 5 mM CMP-sialic acid, at 32.degree. C. for 24 hr in a buffer of 50 mM Tris pH 7.4, 150 mM NaCl, and 3 mM CMP-SA. An aliquot was removed for glycan analysis, and the remaining products were affinity purified as described above.

Detail Description Paragraph - DETX (1331):

[1644] Sialylation. TPA expressed in mammalian cells will often contain a majority of the glycans terminating in sialic acid, but to ensure complete sialylation, it would be beneficial to first perform an in vitro sialylation. TPA in a suitable buffer (most preferably between pH 5.5 and 9, for example Tris buffered saline, pH 7.2) is incubated with CMP sialic acid and sialyltransferase for a time sufficient to convert any glycans lacking sialic acid to sialylated species. Typical conditions would be 1 mg/mL TPA, 3 mM CMP sialic acid, 0.02 U/mL ST3Gal3, 32.degree. C. for 24 hours. Microbial growth can be halted either by sterile filtration or the inclusion of 0.02% sodium azide. The TPA concentration is most preferably in the range 0.1 mg/mL up to the solubility limit of the peptide. The concentration of CMP-SA should be sufficient for there to be excess over the available sites, and might range from 50 .mu.M up to 50 mM, and the temperature from 2.degree. C. up to 40.degree. C. The time required for complete reaction will depend on the temperature, the relative amounts of enzyme to acceptor substrate, the donor substrate concentration, and the pH. Other sialyltransferases that may be capable of adding sialic acid in 2,3 linkage include ST3Gal4; microbial transferases could also be used.

PGPUB-DOCUMENT-NUMBER: 20040126838

PGPUB-FILING-TYPE: new

DOCUMENT-IDENTIFIER: US 20040126838 A1

TITLE: Follicle stimulating hormone: remodeling and glycoconjugation of FSH

PUBLICATION-DATE: July 1, 2004

INVENTOR-INFORMATION:

| NAME | CITY | STATE | COUNTRY | RULE-47 |
|---|---|---|---|---|
| DeFrees, Shawn | North Wales | PA | US | |
| Zopf, David | Wayne | PA | US | |
| Bayer, Robert | San Diego | CA | US | |
| Bowe, Caryn | Doylestown | PA | US | |
| Hakes, David | Willow Grove | PA | US | |
| Chen, Xi | Lansdale | PA | US | |

APPL-NO: 10/ 410997

DATE FILED: April 9, 2003

RELATED-US-APPL-DATA:

child 10410997 A1 20030409

parent continuation-in-part-of 10360779 20030219 US PENDING

child 10410997 A1 20030409

parent continuation-in-part-of 10360770 20030106 US PENDING

child 10410997 A1 20030409

parent continuation-in-part-of 10287994 20021105 US PENDING

child 10287994 20021105 US

parent continuation-of PCT/US02/32263 20021009 US PENDING

non-provisional-of-provisional 60407527 20020828 US

non-provisional-of-provisional 60404249 20020816 US

non-provisional-of-provisional 60396594 20020717 US

non-provisional-of-provisional 60391777 20020625 US

non-provisional-of-provisional 60387292 20020607 US

non-provisional-of-provisional 60334301 20011128 US

non-provisional-of-provisional 60334233 20011128 US

US-CL-CURRENT: 435/68.1, 530/397

ABSTRACT:

The invention includes methods and compositions for remodeling a peptide molecule, including the addition or deletion of one or more glycosyl groups to a peptide, and/or the addition of a modifying group to a peptide.

CROSS REFERENCE TO RELATED APPLICATIONS

[0001] This application is a continuation-in-part of prior Application No. PCT/US02/32263, filed Oct. 9, 2002; Provisional Patent Application No. 60/448,381, filed Feb. 19, 2003 (converted to non-provisional application, same filing date, serial number not yet assigned); Provisional Patent Application No. 60/438,582, filed Jan. 6, 2003 (converted to non-provisional application, same filing date, serial number not yet assigned); Provisional Patent Application No. 60/407,527, filed Aug. 28, 2002; Provisional Patent Application No. 60/404,249, filed Aug. 16, 2002; Provisional Patent Application No. 60/396,594, filed Jul. 17, 2002; Provisional Patent Application No. 60/391,777, filed Jun. 25, 2002; Provisional Patent Application No. 60/387,292, filed Jun. 7, 2002; Provisional Patent Application No. 60/334,301, filed Nov. 28, 2001; Provisional Patent Application No. 60/334,233, filed Nov. 28, 2001; Provisional Patent Application No. 60/344,692, filed Oct. 19, 2001; and Provisional Patent Application No. 60/328,523, filed Oct. 10, 2001.

---------- KWIC ---------

Detail Description Paragraph - DETX (1060):
[1408] In vitro sialylation was carried out using the G2 glycoform antibody (1 mg/mL) by contacting it with 0.1 U/mg ST3Gal3 or 0.1 U/mg ST6Gal1, 5 mM CMP-sialic acid, at 32.degree. C. for 24 hr in a buffer of 50 mM Tris pH 7.4, 150 mM NaCl, and 3 mM CMP-SA. An aliquot was removed for glycan analysis, and the remaining products were affinity purified as described above.

Detail Description Paragraph - DETX (1332):
[1680] Sialylation. TPA expressed in mammalian cells will often contain a majority of the glycans terminating in sialic acid, but to ensure complete sialylation, it would be beneficial to first perform an in vitro sialylation. TPA in a suitable buffer (most preferably between pH 5.5 and 9, for example Tris buffered saline, pH 7.2) is incubated with CMP sialic acid and sialyltransferase for a time sufficient to convert any glycans lacking sialic acid to sialylated species. Typical conditions would be 1 mg/mL TPA, 3 mM CMP sialic acid, 0.02 U/mL ST3Gal3, 32.degree. C. for 24 hours. Microbial growth can be halted either by sterile filtration or the inclusion of 0.02% sodium azide. The TPA concentration is most preferably in the range 0.1 mg/mL up to the solubility limit of the peptide. The concentration of CMP-SA should be sufficient for there to be excess over the available sites, and might range from 50 .mu.M up to 50 mM, and the temperature from 2.degree. C. up to 40.degree. C. The time required for complete reaction will depend on the temperature, the relative amounts of enzyme to acceptor substrate, the donor substrate concentration, and the pH. Other sialyltransferases that may be capable of adding sialic acid in 2,3 linkage include ST3Gal4; microbial transferases could also be used.

PGPUB-DOCUMENT-NUMBER: 20040122216

PGPUB-FILING-TYPE: new

DOCUMENT-IDENTIFIER: US 20040122216 A1

TITLE: Recombinant tissue protective cytokines and encoding nucleic acids thereof for protection, restoration, and enhancement of responsive cells, tissues, and organs

PUBLICATION-DATE: June 24, 2004

INVENTOR-INFORMATION:

| NAME | CITY | STATE | COUNTRY | RULE-47 |
|---|---|---|---|---|
| Nielsen, Jacob | Copenhagen | CT | DK | |
| Pedersen, Jan Torleif | Bronshoj | NY | DK | |
| Gerwien, Jens | Copenhagen | NY | DK | |
| Bay, Katrine | Copenhagen | | DK | |
| Pedersen, Lars Ostergaard | Copenhagen | | | DK |
| Leist, Marcel | Valby | | DK | |
| Geist, Marie Aavang | Valby | | DK | |
| Kallunki, Pekka | Copenhagen | | DK | |
| Christensen, Soren | Jyllinge | | DK | |
| Sager, Thomas | Smorum | | DK | |
| Brines, Michael | Woodbridge | | US | |
| Cerami, Anthony | Somers | | US | |
| Cerami, Carla | Sleepy Hollow | | US | |

APPL-NO: 10/ 612665

DATE FILED: July 1, 2003

RELATED-US-APPL-DATA:

non-provisional-of-provisional 60392455 20020701 US

non-provisional-of-provisional 60393423 20020703 US

US-CL-CURRENT: 530/351

ABSTRACT:

Methods and compositions are provided for protecting or enhancing a responsive cell, tissue, organ or body part function or viability in vivo, in situ or ex vivo in mammals, including human beings, by systemic or local administration of an erythropoietin receptor activity modulator, such as an recombinant tissue protective cytokine.

---------- KWIC --------

Detail Description Paragraph - DETX (80):

[0187] A non-limiting example of sialylation of a glycopeptide is found in U.S. patent application Ser. No. U.S. 2003/0040037, which discloses methods of sialylation using mammalian or bacterial sialytransferases. Another non-limiting example of methods for sialylation and alteration of sialylation patterns on glycoproteins is found in U.S. patent application Ser. No. U.S. 2002/0160460 A1 and in U.S. Pat. No. 6,399,336 B1. Therein, in vitro methods

for sialylating recombinant glycoproteins are disclosed where a sialic acid donor moiety is combined with a glycoprotein having a galactose or N-acetylgalactosamine acceptor moiety. In such methods a sialyltransferase combined with the acceptor and donor attached a sialic acid to a saccharide.

PGPUB-DOCUMENT-NUMBER: 20040115168

PGPUB-FILING-TYPE: new

DOCUMENT-IDENTIFIER: US 20040115168 A1

TITLE: Interferon beta: remodeling and glycoconjugation of interferon beta

PUBLICATION-DATE: June 17, 2004

INVENTOR-INFORMATION:

| NAME | CITY | STATE | COUNTRY | RULE-47 |
|---|---|---|---|---|
| DeFrees, Shawn | North Wales | PA | US | |
| Zopf, David | Wayne | PA | US | |
| Bayer, Robert | San Diego | CA | US | |
| Bowe, Caryn | Doylestown | PA | US | |
| Hakes, David | Willow Grove | PA | US | |
| Chen, Xi | Lansdale | PA | US | |

APPL-NO: 10/ 410930

DATE FILED: April 9, 2003

RELATED-US-APPL-DATA:

child 10410930 A1 20030409

parent continuation-in-part-of 10360779 20030219 US PENDING

child 10410930 A1 20030409

parent continuation-in-part-of 10360770 20030106 US PENDING

child 10410930 A1 20030409

parent continuation-in-part-of 10287994 20021105 US PENDING

child 10287994 20021105 US

parent continuation-of PCT/US02/32263 20021009 US PENDING

non-provisional-of-provisional 60407527 20020828 US

non-provisional-of-provisional 60404249 20020816 US

non-provisional-of-provisional 60396594 20020717 US

non-provisional-of-provisional 60391777 20020625 US

non-provisional-of-provisional 60387292 20020607 US

non-provisional-of-provisional 60334301 20011128 US

non-provisional-of-provisional 60334233 20011128 US

non-provisional-of-provisional 60344692 20011019 US

non-provisional-of-provisional 60328523 20011010 US

US-CL-CURRENT: 424/85.6, 435/68.1 , 530/351

ABSTRACT:

The invention includes methods and compositions for remodeling a peptide molecule, including the addition or deletion of one or more glycosyl groups to a peptide, and/or the addition of a modifying group to a peptide.

CROSS REFERENCE TO RELATED APPLICATIONS

[0001] This application is a continuation-in-part of prior Application No. PCT/US02/32263, filed Oct. 9, 2002; Provisional Patent Application No. 60/448,381, filed Feb. 19, 2003 (converted to non-provisional application, same filing date, serial number not yet assigned); Provisional Patent Application No. 60/438,582, filed Jan. 6, 2003 (converted to non-provisional application, same filing date, serial number not yet assigned); Provisional Patent Application No. 60/407,527, filed Aug. 28, 2002; Provisional Patent Application No. 60/404,249, filed Aug. 16, 2002; Provisional Patent Application No. 60/396,594, filed Jul. 17, 2002; Provisional Patent Application No. 60/391,777, filed Jun. 25, 2002; Provisional Patent Application No. 60/387,292, filed Jun. 7, 2002; Provisional Patent Application No. 60/334,301, filed Nov. 28, 2001; Provisional Patent Application No. 60/334,233, filed Nov. 28, 2001; Provisional Patent Application No. 60/344,692, filed Oct. 19, 2001; and Provisional Patent Application No. 60/328,523, filed Oct. 10, 2001.

---------- KWIC ---------

Detail Description Paragraph - DETX (1056):
[1424] In vitro sialylation was carried out using the G2 glycoform antibody (1 mg/mL) by contacting it with 0.1 U/mg ST3Gal3 or 0.1 U/mg ST6Gal1, 5 mM CMP-sialic acid, at 32.degree. C. for 24 hr in a buffer of 50 mM Tris pH 7.4, 150 mM NaCl, and 3 mM CMP-SA. An aliquot was removed for glycan analysis, and the remaining products were affinity purified as described above.

Detail Description Paragraph - DETX (1325):
[1693] Sialylation. TPA expressed in mammalian cells will often contain a majority of the glycans terminating in sialic acid, but to ensure complete sialylation, it would be beneficial to first perform an in vitro sialylation. TPA in a suitable buffer (most preferably between pH 5.5 and 9, for example Tris buffered saline, pH 7.2) is incubated with CMP sialic acid and sialyltransferase for a time sufficient to convert any glycans lacking sialic acid to sialylated species. Typical conditions would be 1 mg/mL TPA, 3 mM CMP sialic acid, 0.02 U/mL ST3Gal3, 32.degree. C. for 24 hours. Microbial growth can be halted either by sterile filtration or the inclusion of 0.02% sodium azide. The TPA concentration is most preferably in the range 0.1 mg/mL up to the solubility limit of the peptide. The concentration of CMP-SA should be sufficient for there to be excess over the available sites, and might range from 50 .mu.M up to 50 mM, and the temperature from 2.degree. C. up to 40.degree. C. The time required for complete reaction will depend on the temperature, the relative amounts of enzyme to acceptor substrate, the donor substrate concentration, and the pH. Other sialyltransferases that may be capable of adding sialic acid in 2,3 linkage include ST3Gal4; microbial transferases could also be used.

PGPUB-DOCUMENT-NUMBER: 20040110679

PGPUB-FILING-TYPE: new

DOCUMENT-IDENTIFIER: US 20040110679 A1

TITLE: Treatment of disturbances of iron distribution

PUBLICATION-DATE: June 10, 2004

INVENTOR-INFORMATION:

| NAME | CITY | STATE | COUNTRY | RULE-47 |
|---|---|---|---|---|
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APPL-NO: 10/ 634477

DATE FILED: August 4, 2003

FOREIGN-APPL-PRIORITY-DATA:

| COUNTRY | APPL-NO | DOC-ID | APPL-DATE |
|---|---|---|---|
| EP | 02019100.3 | 2002EP-02019100.3 | August 29, 2002 |

US-CL-CURRENT: 514/12

ABSTRACT:

A method of, and pharmaceutical composition for, treating disturbances of iron distribution in diabetes using erythropoietin are disclosed.

---------- KWIC ---------

Detail Description Paragraph - DETX (4):

[0017] Further, erythropoietin may be a glycoprotein analog having from 1 to 6 additional sites for glycosylation. Therefore, the present invention also relates to the use as described before, wherein the erythropoietin protein has the sequence of human erythropoietin modified by the addition of from 1 to 6 glycosylation sites. Glycosylation of a protein, with one or more oligosaccharide groups, occurs at specific locations along a polypeptide backbone and greatly affects the physical properties of the protein such as protein stability, secretion, subcellular localization, and biological activity. Glycosylation is usually of two types. O-linked oligosaccharides are attached to serine or threonine residues and N-linked oligosaccharides are attached to asparagine residues. One type of oligosaccharide found on both N-linked and O-linked oligosaccharides is N-acetylneuraminic acid (sialic acid), which is a family of amino sugars containing 9 or more carbon atoms. Sialic acid is usually the terminal residue on both N-linked and O-linked oligosaccharides and, because it bears a negative charge, confers acidic properties to the glycoprotein. Human erythropoietin, having 165 amino acids, contains three N-linked and one O-linked oligosaccharide chains which comprise about 40% of the total molecular weight of the glycoprotein. N-linked glycosylation occurs at asparagine residues located at positions 24, 38, and 83 and O-linked glycosylation occurs at a serine residue located at position 126. The oligosaccharide chains are modified with terminal sialic acid residues. Enzymatic removal of all sialic acid residues from the glycosylated erythropoietin results in loss of in vivo activity but not in vitro activity

because sialylation of erythropoietin prevents its binding, and subsequent clearance, by hepatic binding protein.

PGPUB-DOCUMENT-NUMBER: 20040082026

PGPUB-FILING-TYPE: new

DOCUMENT-IDENTIFIER: US 20040082026 A1

TITLE: Interferon alpha: remodeling and glycoconjugation of interferon alpha

PUBLICATION-DATE: April 29, 2004

INVENTOR-INFORMATION:

| NAME | CITY | STATE | COUNTRY | RULE-47 |
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| Bayer, Robert | San Diego | CA | US | |
| Bowe, Caryn | Doylestown | PA | US | |
| Hakes, David | Willow Grove | PA | US | |
| Chen, Xi | Lansdale | PA | US | |

APPL-NO: 10/ 411049

DATE FILED: April 9, 2003

RELATED-US-APPL-DATA:

child 10411049 A1 20030409

parent continuation-in-part-of 10360779 20030219 US PENDING

child 10411049 A1 20030409

parent continuation-in-part-of 10360770 20030106 US PENDING

child 10411049 A1 20030409

parent continuation-in-part-of 10287994 20021105 US PENDING

child 10287994 20021105 US

parent continuation-of PCT/US02/32263 20021009 US PENDING

non-provisional-of-provisional 60407527 20020828 US

non-provisional-of-provisional 60404249 20020816 US

non-provisional-of-provisional 60396594 20020717 US

non-provisional-of-provisional 60391777 20020625 US

non-provisional-of-provisional 60387292 20020607 US

non-provisional-of-provisional 60334301 20011128 US

non-provisional-of-provisional 60334233 20011128 US

non-provisional-of-provisional 60344692 20011019 US

non-provisional-of-provisional 60328523 20011010 US

US-CL-CURRENT: 435/68.1, 530/351

ABSTRACT:

The invention includes a multitude of methods and compositions for remodeling a peptide molecule, including the addition or deletion of one or more glycosyl groups to a peptide, and/or the addition of a modifying group to a peptide.

CROSS REFERENCE TO RELATED APPLICATIONS

[0001] This application is a continuation-in-part of prior Application No. PCT/US02/32263, filed Oct. 9, 2002; Provisional Patent Application No. 60/448,381, filed Feb. 19, 2003 (converted to non-provisional application, same filing date, serial number not yet assigned); Provisional Patent Application No. 60/438,582, filed Jan. 6, 2003 (converted to non-provisional application, same filing date, serial number not yet assigned); Provisional Patent Application No. 60/407,527, filed Aug. 28, 2002; Provisional Patent Application No. 60/404,249, filed Aug. 16, 2002; Provisional Patent Application No. 60/396,594, filed Jul. 17, 2002; Provisional Patent Application No. 60/391,777, filed Jun. 25, 2002; Provisional Patent Application No. 60/387,292, filed Jun. 7, 2002; Provisional Patent Application No. 60/334,301, filed Nov. 28, 2001; Provisional Patent Application No. 60/334,233, filed Nov. 28, 2001; Provisional Patent Application No. 60/344,692, filed Oct. 19, 2001; and Provisional Patent Application No. 60/328,523, filed Oct. 10, 2001.

---------- KWIC ---------

Detail Description Paragraph - DETX (1062):
[1465] In vitro sialylation was carried out using the G2 glycoform antibody (1 mg/mL) by contacting it with 0.1 U/mg ST3Gal3 or 0.1 U/mg ST6Gal1, 5 mM CMP-sialic acid, at 32.degree. C. for 24 hr in a buffer of 50 mM Tris pH 7.4, 150 mM NaCl, and 3 mM CMP-SA. An aliquot was removed for glycan analysis, and the remaining products were affinity purified as described above.

Detail Description Paragraph - DETX (1334):
[1737] Sialylation. TPA expressed in mammalian cells will often contain a majority of the glycans terminating in sialic acid, but to ensure complete sialylation, it would be beneficial to first perform an in vitro sialylation. TPA in a suitable buffer (most preferably between pH 5.5 and 9, for example Tris buffered saline, pH 7.2) is incubated with CMP sialic acid and sialyltransferase for a time sufficient to convert any glycans lacking sialic acid to sialylated species. Typical conditions would be 1 mg/mL TPA, 3 mM CMP sialic acid, 0.02 U/mL ST3Gal3, 32.degree. C. for 24 hours. Microbial growth can be halted either by sterile filtration or the inclusion of 0.02% sodium azide. The TPA concentration is most preferably in the range 0.1 mg/mL up to the solubility limit of the peptide. The concentration of CMP-SA should be sufficient for there to be excess over the available sites, and might range from 50 .mu.M up to 50 mM, and the temperature from 2.degree. C. up to 40.degree. C. The time required for complete reaction will depend on the temperature, the relative amounts of enzyme to acceptor substrate, the donor substrate concentration, and the pH. Other sialyltransferases that may be capable of adding sialic acid in 2,3 linkage include ST3Gal4; microbial transferases could also be used.

PGPUB-DOCUMENT-NUMBER: 20040077836

PGPUB-FILING-TYPE: new

DOCUMENT-IDENTIFIER: US 20040077836 A1

TITLE: Granulocyte colony stimulating factor: remodeling and glycoconjugation of G-CSF

PUBLICATION-DATE: April 22, 2004

INVENTOR-INFORMATION:

| NAME | CITY | STATE | COUNTRY | RULE-47 |
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| Zopf, David | Wayne | PA | US | |
| Bayer, Robert | San Diego | CA | US | |
| Bowe, Caryn | Doylestown | PA | US | |
| Hakes, David | Willow Grove | PA | US | |
| Chen, Xi | Lansdale | PA | US | |

APPL-NO: 10/ 410962

DATE FILED: April 9, 2003

RELATED-US-APPL-DATA:

child 10410962 A1 20030409

parent continuation-in-part-of 10360779 20030219 US PENDING

child 10410962 A1 20030409

parent continuation-in-part-of 10360770 20030106 US PENDING

child 10360770 20030106 US

parent continuation-in-part-of 10287994 20021105 US PENDING

child 10287994 20021105 US

parent continuation-of PCT/US02/32263 20021009 US PENDING

non-provisional-of-provisional 60407527 20020828 US

non-provisional-of-provisional 60404249 20020816 US

non-provisional-of-provisional 60396594 20020717 US

non-provisional-of-provisional 60391777 20020625 US

non-provisional-of-provisional 60387292 20020607 US

non-provisional-of-provisional 60334301 20011128 US

non-provisional-of-provisional 60334233 20011128 US

non-provisional-of-provisional 60344692 20011019 US

non-provisional-of-provisional 60328523 20011010 US

US-CL-CURRENT: 530/351, 435/68.1

ABSTRACT:

The invention includes methods and compositions for remodeling a peptide molecule, including the addition or deletion of one or more glycosyl groups to a peptide, and/or the addition of a modifying group to a peptide.

CROSS REFERENCE TO RELATED APPLICATIONS

[0001] This application is a continuation-in-part of prior Application No. PCT/US02/32263, filed Oct. 9, 2002; Provisional Patent Application No. 60/448,381, filed Feb. 19, 2003 (converted to non-provisional application, same filing date, serial number not yet assigned); Provisional Patent Application No. 60/438,582, filed Jan. 6, 2003 (converted to non-provisional application, same filing date, serial number not yet assigned); Provisional Patent Application No. 60/407,527, filed Aug. 28, 2002; Provisional Patent Application No. 60/404,249, filed Aug. 16, 2002; Provisional Patent Application No. 60/396,594, filed Jul. 17, 2002; Provisional Patent Application No. 60/391,777, filed Jun. 25, 2002; Provisional Patent Application No. 60/387,292, filed Jun. 7, 2002; Provisional Patent Application No. 60/334,301, filed Nov. 28, 2001; Provisional Patent Application No. 60/334,233, filed Nov. 28, 2001; Provisional Patent Application No. 60/344,692, filed Oct. 19, 2001; and Provisional Patent Application No. 60/328,523, filed Oct. 10, 2001.

---------- KWIC ---------

Detail Description Paragraph - DETX (1057):
[1392] In vitro sialylation was carried out using the G2 glycoform antibody (1 mg/mL) by contacting it with 0.1 U/mg ST3Gal3 or 0.1 U/mg ST6Gal 1, 5 mM CMP-sialic acid, at 32.degree. C. for 24 hr in a buffer of 50 mM Tris pH 7.4, 150 mM NaCl, and 3 mM CMP-SA. An aliquot was removed for glycan analysis, and the remaining products were affinity purified as described above.

Detail Description Paragraph - DETX (1328):
[1663] Sialylation. TPA expressed in mammalian cells will often contain a majority of the glycans terminating in sialic acid, but to ensure complete sialylation, it would be beneficial to first perform an in vitro sialylation. TPA in a suitable buffer (most preferably between pH 5.5 and 9, for example Tris buffered saline, pH 7.2) is incubated with CMP sialic acid and sialyltransferase for a time sufficient to convert any glycans lacking sialic acid to sialylated species. Typical conditions would be 1 mg/mL TPA, 3 mM CMP sialic acid, 0.02 U/mL ST3Gal3, 32.degree. C. for 24 hours. Microbial growth can be halted either by sterile filtration or the inclusion of 0.02% sodium azide. The TPA concentration is most preferably in the range 0.1 mg/mL up to the solubility limit of the peptide. The concentration of CMP-SA should be sufficient for there to be excess over the available sites, and might range from 50 .mu.M up to 50 mM, and the temperature from 2.degree. C. up to 40.degree. C. The time required for complete reaction will depend on the temperature, the relative amounts of enzyme to acceptor substrate, the donor substrate concentration, and the pH. Other sialyltransferases that may be capable of adding sialic acid in 2,3 linkage include ST3Gal4; microbial transferases could also be used.

PGPUB-DOCUMENT-NUMBER: 20040063911

PGPUB-FILING-TYPE: new

DOCUMENT-IDENTIFIER: US 20040063911 A1

TITLE: Protein remodeling methods and proteins/peptides produced by the methods

PUBLICATION-DATE: April 1, 2004

INVENTOR-INFORMATION:

| NAME | CITY | STATE | COUNTRY | RULE-47 |
|---|---|---|---|---|
| DeFrees, Shawn | North Wales | PA | US | |
| Zopf, David | Wayne | PA | US | |
| Bayer, Robert | San Diego | CA | US | |
| Hakes, David | Willow Grove | PA | US | |
| Chen, Xi | Lansdale | PA | US | |

APPL-NO: 10/ 411026

DATE FILED: April 9, 2003

RELATED-US-APPL-DATA:

child 10411026 A1 20030409

parent continuation-in-part-of 10360779 20030219 US PENDING

child 10360779 20030219 US

parent continuation-in-part-of 10360770 20030106 US PENDING

child 10360770 20030106 US

parent continuation-in-part-of 10287994 20021105 US PENDING

child 10287994 20021105 US

parent continuation-of PCT/US02/32263 20021009 US PENDING

non-provisional-of-provisional 60407527 20020828 US

non-provisional-of-provisional 60404249 20020816 US

non-provisional-of-provisional 60396594 20020717 US

non-provisional-of-provisional 60391777 20020625 US

non-provisional-of-provisional 60387292 20020607 US

non-provisional-of-provisional 60334301 20011128 US

non-provisional-of-provisional 60334233 20011128 US

non-provisional-of-provisional 60344692 20011019 US

non-provisional-of-provisional 60328523 20011010 US

US-CL-CURRENT: 530/351, 435/68.1 , 530/395

ABSTRACT:

The invention includes methods and compositions for remodeling a peptide molecule, including the addition or deletion of one or more glycosyl groups to a peptide, and/or the addition of a modifying group to a peptide.

CROSS REFERENCE TO RELATED APPLICATIONS

[0001] This application is a continuation-in-part of prior Application No. PCT/US02/32263, filed Oct. 9, 2002; Provisional Patent Application No. 60/448,381, filed Feb. 19, 2003 (converted to non-provisional application, same filing date, serial number not yet assigned); Provisional Patent Application No. 60/438,582, filed Jan. 6, 2003 (converted to non-provisional application, same filing date, serial number not yet assigned); Provisional Patent Application No. 60/407,527, filed Aug. 28, 2002; Provisional Patent Application No. 60/404,249, filed Aug. 16, 2002; Provisional Patent Application No. 60/396,594, filed Jul. 17, 2002; Provisional Patent Application No. 60/391,777, filed Jun. 25, 2002; Provisional Patent Application No. 60/387,292, filed Jun. 7, 2002; Provisional Patent Application No. 60/334,301, filed Nov. 28, 2001; Provisional Patent Application No. 60/334,233, filed Nov. 28, 2001; Provisional Patent Application No. 60/344,692, filed Oct. 19, 2001; and Provisional Patent Application No. 60/328,523, filed Oct. 10, 2001.

---------- KWIC ---------

Detail Description Paragraph - DETX (1060):
[1341] In vitro sialylation was carried out using the G2 glycoform antibody (1 mg/mL) by contacting it with 0.1 U/mg ST3Gal3 or 0.1 U/mg ST6Gal 1, 5 mM CMP-sialic acid, at 32.degree. C. for 24 hr in a buffer of 50 mM Tris pH 7.4, 150 mM NaCl, and 3 mM CMP-SA. An aliquot was removed for glycan analysis, and the remaining products were affinity purified as described above.

Detail Description Paragraph - DETX (1334):
[1615] Sialylation. TPA expressed in mammalian cells will often contain a majority of the glycans terminating in sialic acid, but to ensure complete sialylation, it would be beneficial to first perform an in vitro sialylation. TPA in a suitable buffer (most preferably between pH 5.5 and 9, for example Tris buffered saline, pH 7.2) is incubated with CMP sialic acid and sialyltransferase for a time sufficient to convert any glycans lacking sialic acid to sialylated species. Typical conditions would be 1 mg/mL TPA, 3 mM CMP sialic acid, 0.02 U/mL ST3Gal3, 32.degree. C. for 24 hours. Microbial growth can be halted either by sterile filtration or the inclusion of 0.02% sodium azide. The TPA concentration is most preferably in the range 0.1 mg/mL up to the solubility limit of the peptide. The concentration of CMP-SA should be sufficient for there to be excess over the available sites, and might range from 50 .mu.M up to 50 mM, and the temperature from 2.degree. C. up to 40.degree. C. The time required for complete reaction will depend on the temperature, the relative amounts of enzyme to acceptor substrate, the donor substrate concentration, and the pH. Other sialyltransferases that may be capable of adding sialic acid in 2,3 linkage include ST3Gal4; microbial transferases could also be used.

PGPUB-DOCUMENT-NUMBER: 20040043446

PGPUB-FILING-TYPE: new

DOCUMENT-IDENTIFIER: US 20040043446 A1

TITLE: Alpha galalctosidase a: remodeling and glycoconjugation of alpha galactosidase A

PUBLICATION-DATE: March 4, 2004

INVENTOR-INFORMATION:

| NAME | CITY | STATE | COUNTRY | RULE-47 |
|---|---|---|---|---|
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| Zopf, David | Wayne | PA | US | |
| Bayer, Robert | San Diego | CA | US | |
| Bowe, Caryn | Doylestown | PA | US | |
| Hakes, David | Willow Grove | PA | US | |
| Chen, Xi | Lansdale | PA | US | |

APPL-NO: 10/ 411037

DATE FILED: April 9, 2003

RELATED-US-APPL-DATA:

child 10411037 A1 20030409

parent continuation-in-part-of PCT/US02/32263 20021009 US PENDING

non-provisional-of-provisional 60407527 20020828 US

non-provisional-of-provisional 60404249 20020816 US

non-provisional-of-provisional 60396594 20020717 US

non-provisional-of-provisional 60391777 20020625 US

non-provisional-of-provisional 60387292 20020607 US

US-CL-CURRENT: 435/68.1, 435/193 , 435/208

ABSTRACT:

The invention includes methods and compositions for remodeling a peptide molecule, including the addition or deletion of one or more glycosyl groups to a peptide, and/or the addition of a modifying group to a peptide.

CROSS REFERENCE TO RELATED APPLICATIONS

[0001] This application is a continuation-in-part of prior Application No. PCT/US02/32263, filed Oct. 9, 2002; Provisional Patent Application No. 60/448,381, filed Feb. 19, 2003 (converted to non-provisional application, same filing date, serial number not yet assigned); Provisional Patent Application No. 60/438,582, filed Jan. 6, 2003 (converted to non-provisional application, same filing date, serial number not yet assigned); Provisional Patent Application No. 60/407,527, filed Aug. 28, 2002; Provisional Patent Application No. 60/404,249, filed Aug. 16, 2002; Provisional Patent

Application No. 60/396,594, filed Jul. 17, 2002; Provisional Patent Application No. 60/391,777, filed Jun. 25, 2002; Provisional Patent Application No. 60/387,292, filed Jun. 7, 2002; Provisional Patent Application No. 60/334,301, filed Nov. 28, 2001; Provisional Patent Application No. 60/334,233, filed Nov. 28, 2001; Provisional Patent Application No. 60/344,692, filed Oct. 19, 2001; and Provisional Patent Application No. 60/328,523, filed Oct. 10, 2001.

---------- KWIC ---------

Detail Description Paragraph - DETX (1049):

[1413] In vitro sialylation was carried out using the G2 glycoform antibody (1 mg/mL) by contacting it with 0.1 U/mg ST3Ga3 or 0.1 U/mg ST6Gal1, 5 mM CMP-sialic acid, at 32.degree. C. for 24 hr in a buffer of 50 mM Tris pH 7.4, 150 mM NaCl, and 3 mM CMP-SA. An aliquot was removed for glycan analysis, and the remaining products were affinity purified as described above.

Detail Description Paragraph - DETX (1318):

[1682] Sialylation. TPA expressed in mammalian cells will often contain a majority of the glycans terminating in sialic acid, but to ensure complete sialylation, it would be beneficial to first perform an in vitro sialylation. TPA in a suitable buffer (most preferably between pH 5.5 and 9, for example Tris buffered saline, pH 7.2) is incubated with CMP sialic acid and sialyltransferase for a time sufficient to convert any glycans lacking sialic acid to sialylated species. Typical conditions would be 1 mg/mL TPA, 3 mM CMP sialic acid, 0.02 U/mL ST3Gal3, 32.degree. C. for 24 hours. Microbial growth can be halted either by sterile filtration or the inclusion of 0.02% sodium azide. The TPA concentration is most preferably in the range 0.1 mg/mL up to the solubility limit of the peptide. The concentration of CMP-SA should be sufficient for there to be excess over the available sites, and might range from 50 .mu.M up to 50 mM, and the temperature from 2.degree. C. up to 40.degree. C. The time required for complete reaction will depend on the temperature, the relative amounts of enzyme to acceptor substrate, the donor substrate concentration, and the pH. Other sialyltransferases that may be capable of adding sialic acid in 2,3 linkage include ST3Gal4; microbial transferases could also be used.

PGPUB-DOCUMENT-NUMBER: 20040018590

PGPUB-FILING-TYPE: new

DOCUMENT-IDENTIFIER: US 20040018590 A1

TITLE: Combinatorial DNA library for producing modified N-glycans in lower eukaryotes

PUBLICATION-DATE: January 29, 2004

INVENTOR-INFORMATION:

| NAME | CITY | STATE | COUNTRY | RULE-47 |
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| Gerngross, Tillman U. | Hanover | NH | US | |
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| Choi, Byung-Kwon | Norwich | VT | US | |
| Nett, Juergen Hermann | Grantham | NH | US | |
| Bobrowicz, Piotr | White River Junction | VT | US | |
| Hamilton, Stephen R. | Enfield | NH | US | |
| Davidson, Robert C. | Enfield | NH | US | |


APPL-NO: 10/ 371877

DATE FILED: February 20, 2003

RELATED-US-APPL-DATA:

child 10371877 A1 20030220

parent continuation-in-part-of 09892591 20010627 US PENDING

non-provisional-of-provisional 60214358 20000628 US

non-provisional-of-provisional 60215638 20000630 US

non-provisional-of-provisional 60279997 20010330 US

US-CL-CURRENT: 435/69.1, 435/320.1 , 435/325 , 530/395 , 536/23.2 , 536/53

ABSTRACT:

The present invention relates to eukaryotic host cells having modified oligosaccharides which may be modified further by heterologous expression of a set of glycosyltransferases, sugar transporters and mannosidases to become host-strains for the production of mammalian, e.g., human therapeutic glycoproteins. The invention provides nucleic acid molecules and combinatorial libraries which can be used to successfully target and express mammalian enzymatic activities such as those involved in glycosylation to intracellular compartments in a eukaryotic host cell. The process provides an engineered host cell which can be used to express and target any desirable gene(s) involved in glycosylation. Host cells with modified oligosaccharides are created or selected. N-glycans made in the engineered host cells have a Man.sub.5GlcNAc.sub.2 core structure which may then be modified further by heterologous expression of one or more enzymes, e.g., glycosyltransferases, sugar transporters and mannosidases, to yield human-like glycoproteins. For the production of therapeutic proteins, this method may be adapted to engineer cell lines in which any desired glycosylation structure may be obtained.

CROSS-REFERENCE TO RELATED APPLICATIONS

[0001] This application is a continuation-in-part of U.S. application Ser. No. 09/892,591, filed Jun. 27, 2001, in which priority is claimed to U.S. Provisional Application Serial No. 60/214,358, filed Jun. 28, 2000, U.S. Provisional Application No. 60/215,638, filed Jun. 30, 2000, and U.S. Provisional Application No. 60/279,997, filed Mar. 30, 2001; each of which is incorporated herein by reference in its entirety.

---------- KWIC ---------

Summary of Invention Paragraph - BSTX (19):

[0014] A significant number of proteins isolated from humans or animals are post-translationally modified, with glycosylation being one of the most significant modifications. An estimated 70% of all therapeutic proteins are glycosylated and thus currently rely on a production system (i.e., host cell) that is able to glycosylate in a manner similar to humans. Several studies have shown that glycosylation plays an important role in determining the (1) immunogenicity, (2) pharmacokinetic properties, (3) trafficking, and (4) efficacy of therapeutic proteins. It is thus not surprising that substantial efforts by the pharmaceutical industry have been directed at developing processes to obtain glycoproteins that are as "humanoid" or "human-like" as possible. To date, most glycoproteins are made in a mammalian host system. This may involve the genetic engineering of such mammalian cells to enhance the degree of sialylation (i.e., terminal addition of sialic acid) of proteins expressed by the cells, which is known to improve pharmacokinetic properties of such proteins. Alternatively, one may improve the degree of sialylation by in vitro addition of such sugars using known glycosyltransferases and their respective nucleotide sugars (e.g., 2,3-sialyltransferase and CMP-sialic acid).

PGPUB-DOCUMENT-NUMBER: 20040016005

PGPUB-FILING-TYPE: new

DOCUMENT-IDENTIFIER: US 20040016005 A1

TITLE: Production of butyrylcholinesterases in transgenic mammals

PUBLICATION-DATE: January 22, 2004

INVENTOR-INFORMATION:

| NAME | CITY | STATE | COUNTRY | RULE-47 |
|---|---|---|---|---|
| Karatzas, Costas N. | Quebec | | CA | |
| Huang, Yue-Jin | Quebec | | CA | |
| Lazaris, Anthoula | Quebec | | CA | |


APPL-NO: 10/ 326892

DATE FILED: December 20, 2002

RELATED-US-APPL-DATA:

non-provisional-of-provisional 60344295 20011221 US

US-CL-CURRENT: 800/7, 800/14 , 800/18

ABSTRACT:

The present invention provides methods for the large-scale production of recombinant butyrylcholinesterase in cell culture, and in the milk and/or urine of transgenic mammals. The recombinant butyrylcholinesterases of this invention can be used to treat and/or prevent organophosphate pesticide poisoning, nerve gas poisoning, cocaine intoxication, and succinylcholine-induced apnea.

[0001] This application claims priority to provisional U.S. application No. 60/344,295 filed Dec. 21, 2001 under 35 U.S.C. .sctn. 119(e), which is incorporated by reference.

---------- KWIC ---------

Detail Description Paragraph - DETX (33):

[0083] As a means of producing recombinant BChE with a glycosylation profile that more closely resembles that of the native enzyme, the present invention is directed to transgenic animals that express both a BChE enzyme and one or more glycosyltransferases in their mammary glands and/or urinary endothelium, as well as cultured mammalian cells that express both a BChE enzyme and one or more glycosyltransferases. The presence of the glycosyltransferases in the intracellular secretory pathway of cells that are also expressing a secreted form of BChE catalyzes the transfer of glycan moieties to said BChE enzymes. The invention also encompasses addition of one or more glycosyltransferases to an in vitro reaction for the transfer of glycan moieties to a recombinant BChE produced by the transgenic animals or transfected mammalian cell lines of the invention. For example, recombinant BChE may be sialylated using the in vitro

reaction conditions described in Chitlaru, et al. Biochem. J. (1998) 336:647-658. Thus, the glycosyltransferase which catalyzes transfer of glycans to the BChE enzyme may be expressed by the same cell that expresses the BChE enzyme, or the glycosyltransferase may be obtained from an external source and added to the recombinant BChE.

| Ref # | Hits | Search Query | DBs | Default Operator | Plurals | Time Stamp |
|---|---|---|---|---|---|---|
| L1 | 32 | vitro near4 sialyla$ | US-PGPUB; USPAT | OR | OFF | 2004/07/12 14:11 |
| L2 | 38 | (commercial or scale or batch) near4 sialyl$ | US-PGPUB; USPAT | OR | OFF | 2004/07/12 14:20 |

PGPUB-DOCUMENT-NUMBER: 20040132640

PGPUB-FILING-TYPE: new

DOCUMENT-IDENTIFIER: US 20040132640 A1

TITLE: Glycopegylation methods and proteins/peptides produced by the methods

PUBLICATION-DATE: July 8, 2004

INVENTOR-INFORMATION:

| NAME | CITY | STATE | COUNTRY | RULE-47 |
|---|---|---|---|---|
| DeFrees, Shawn | North Wales | PA | US | |
| Zopf, David | Wayne | PA | US | |
| Bayer, Robert | San Diego | CA | US | |
| Bowe, Caryn | Doylestown | PA | US | |
| Hakes, David | Willow Grove | PA | US | |
| Chen, Xi | Lansdale | PA | US | |

APPL-NO: 10/ 411012

DATE FILED: April 9, 2003

RELATED-US-APPL-DATA:

child 10411012 A1 20030409

parent continuation-in-part-of PCT/US02/32263 20021009 US PENDING

non-provisional-of-provisional 60407527 20020828 US

non-provisional-of-provisional 60404249 20020816 US

non-provisional-of-provisional 60396594 20020717 US

non-provisional-of-provisional 60391777 20020625 US

non-provisional-of-provisional 60387292 20020607 US

US-CL-CURRENT: 514/8, 530/395

ABSTRACT:

The invention includes methods and compositions for remodeling a peptide molecule, including the addition or deletion of one or more glycosyl groups to a peptide, and/or the addition of a modifying group to a peptide.

CROSS REFERENCE TO RELATED APPLICATIONS

[0001] This application is a continuation-in-part of prior Application No. PCT/US02/32263, filed Oct. 9, 2002; Provisional Patent Application No. 60/448,381, filed Feb. 19, 2003 (converted to non-provisional application, same filing date, serial number not yet assigned); Provisional Patent Application No. 60/438,582, filed Jan. 6, 2003 (converted to non-provisional application, same filing date, serial number not yet assigned); Provisional Patent Application No. 60/407,527, filed Aug. 28, 2002; Provisional Patent Application No. 60/404,249, filed Aug. 16, 2002; Provisional Patent

Application No. 60/396,594, filed Jul. 17, 2002; Provisional Patent Application No. 60/391,777, filed Jun. 25, 2002; Provisional Patent Application No. 60/387,292, filed Jun. 7, 2002; Provisional Patent Application No. 60/334,301, filed Nov. 28, 2001; Provisional Patent Application No. 60/334,233, filed Nov. 28, 2001; Provisional Patent Application No. 60/344,692, filed Oct. 19, 2001; and Provisional Patent Application No. 60/328,523, filed Oct. 10, 2001.

---------- KWIC ---------

Detail Description Paragraph - DETX (669):

[0983] Other sialyltransferases, including those listed in Table 8, are also useful in an economic and efficient large-scale process for sialylation of commercially important glycopeptides. As a simple test to find out the utility of these other enzymes, various amounts of each enzyme (1-100 mU/mg protein) are reacted with asialo-al AGP (at 1-10 mg/ml) to compare the ability of the sialyltransferase of interest to sialylate glycopeptides relative to either bovine ST6Gal I, ST3Gal III or both sialyltransferases. Alternatively, other glycopeptides or glycopeptides, or N-linked oligosaccharides enzymatically released from the peptide backbone can be used in place of asialo-.alpha..sub.1 AGP for this evaluation. Sialyltransferases with the ability to sialylate N-linked oligosaccharides of glycopeptides more efficiently than ST6Gal I are useful in a practical large-scale process for peptide sialylation (as illustrated for ST3Gal III in this disclosure).

Detail Description Paragraph - DETX (1046):

[1359] This Example demonstrates that sialylation of recombinant glycoproteins using the ST3 Gal III sialyltransferase required much less enzyme than expected. For a one kilogram scale reaction, approximately 7,000 units of the ST3Gal III sialyltransferase would be needed, instead of 100,000-150,000 units that earlier studies indicated. Purification of these enzymes from natural sources is prohibitive, with yields of only 1-10 units for a large scale preparation after 1-2 months work. Assuming that both the ST6Gal I and ST3Gal III sialyltransferases are produced as recombinant sialyltransferases, with equal levels of expression of the two enzymes being achieved, a fermentation scale 14-21 times greater (or more) would be required for the ST6Gal I sialyltransferase relative to the ST3Gal III sialyltransferase. For the ST6Gal I sialyltransferase, expression levels of 0.3 U/l in yeast has been reported (Borsig et al. (1995) Biochem. Biophys. Res. Commun. 210: 14-20). Expression levels of 1000 U/liter of the ST3 Gal III sialyltransferase have been achieved in Aspergillus niger. At current levels of expression 300-450,000 liters of yeast fermentation would be required to produce sufficient enzyme for sialylation of 1 kg of glycoprotein using the ST6Gal I sialyltransferase. In contrast, less than 10 liter fermentation of Aspergillus niger would be required for sialylation of 1 kg of glycoprotein using the ST3Gal III sialyltransferase. Thus, the fermentation capacity required to produce the ST3Gal III sialyltransferase for a large scale sialylation reaction would be 10-100 fold less than that required for producing the ST6Gal I; the cost of producing the sialyltransferase would be reduced proportionately.

PGPUB-DOCUMENT-NUMBER: 20040126838

PGPUB-FILING-TYPE: new

DOCUMENT-IDENTIFIER: US 20040126838 A1

TITLE: Follicle stimulating hormone: remodeling and glycoconjugation of FSH

PUBLICATION-DATE: July 1, 2004

INVENTOR-INFORMATION:

| NAME | CITY | STATE | COUNTRY | RULE-47 |
|---|---|---|---|---|
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| Zopf, David | Wayne | PA | US | |
| Bayer, Robert | San Diego | CA | US | |
| Bowe, Caryn | Doylestown | PA | US | |
| Hakes, David | Willow Grove | PA | US | |
| Chen, Xi | Lansdale | PA | US | |

APPL-NO: 10/ 410997

DATE FILED: April 9, 2003

RELATED-US-APPL-DATA:

child 10410997 A1 20030409

parent continuation-in-part-of 10360779 20030219 US PENDING

child 10410997 A1 20030409

parent continuation-in-part-of 10360770 20030106 US PENDING

child 10410997 A1 20030409

parent continuation-in-part-of 10287994 20021105 US PENDING

child 10287994 20021105 US

parent continuation-of PCT/US02/32263 20021009 US PENDING

non-provisional-of-provisional 60407527 20020828 US

non-provisional-of-provisional 60404249 20020816 US

non-provisional-of-provisional 60396594 20020717 US

non-provisional-of-provisional 60391777 20020625 US

non-provisional-of-provisional 60387292 20020607 US

non-provisional-of-provisional 60334301 20011128 US

non-provisional-of-provisional 60334233 20011128 US

US-CL-CURRENT: 435/68.1, 530/397

ABSTRACT:

The invention includes methods and compositions for remodeling a peptide molecule, including the addition or deletion of one or more glycosyl groups to a peptide, and/or the addition of a modifying group to a peptide.

CROSS REFERENCE TO RELATED APPLICATIONS

[0001] This application is a continuation-in-part of prior Application No. PCT/US02/32263, filed Oct. 9, 2002; Provisional Patent Application No. 60/448,381, filed Feb. 19, 2003 (converted to non-provisional application, same filing date, serial number not yet assigned); Provisional Patent Application No. 60/438,582, filed Jan. 6, 2003 (converted to non-provisional application, same filing date, serial number not yet assigned); Provisional Patent Application No. 60/407,527, filed Aug. 28, 2002; Provisional Patent Application No. 60/404,249, filed Aug. 16, 2002; Provisional Patent Application No. 60/396,594, filed Jul. 17, 2002; Provisional Patent Application No. 60/391,777, filed Jun. 25, 2002; Provisional Patent Application No. 60/387,292, filed Jun. 7, 2002; Provisional Patent Application No. 60/334,301, filed Nov. 28, 2001; Provisional Patent Application No. 60/334,233, filed Nov. 28, 2001; Provisional Patent Application No. 60/344,692, filed Oct. 19, 2001; and Provisional Patent Application No. 60/328,523, filed Oct. 10, 2001.

---------- KWIC ---------

Detail Description Paragraph - DETX (668):
[1020] Other sialyltransferases, including those listed in Table 8, are also useful in an economic and efficient large-scale process for sialylation of commercially important glycopeptides. As a simple test to find out the utility of these other enzymes, various amounts of each enzyme (1-100 mU/mg protein) are reacted with asialo-.alpha..sub.1 AGP (at 1-10 mg/ml) to compare the ability of the sialyltransferase of interest to sialylate glycopeptides relative to either bovine ST6Gal I, ST3Gal III or both sialyltransferases. Alternatively, other glycopeptides or glycopeptides, or N-linked oligosaccharides enzymatically released from the peptide backbone can be used in place of asialo-.alpha..sub.1 AGP for this evaluation. Sialyltransferases with the ability to sialylate N-linked oligosaccharides of glycopeptides more efficiently than ST6Gal I are useful in a practical large-scale process for peptide sialylation (as illustrated for ST3Gal III in this disclosure).

Detail Description Paragraph - DETX (1047):
[1395] This Example demonstrates that sialylation of recombinant glycoproteins using the ST3 Gal III sialyltransferase required much less enzyme than expected. For a one kilogram scale reaction, approximately 7,000 units of the ST3Gal III sialyltransferase would be needed, instead of 100,000-150,000 units that earlier studies indicated. Purification of these enzymes from natural sources is prohibitive, with yields of only 1-10 units for a large scale preparation after 1-2 months work. Assuming that both the ST6Gal I and ST3Gal III sialyltransferases are produced as recombinant sialyltransferases, with equal levels of expression of the two enzymes being achieved, a fermentation scale 14-21 times greater (or more) would be required for the ST6Gal I sialyltransferase relative to the ST3Gal III sialyltransferase. For the ST6Gal I sialyltransferase, expression levels of 0.3 U/1 in yeast has been reported (Borsig et al. (1995) Biochem. Biophys. Res. Commun. 210: 14-20). Expression levels of 1000 U/liter of the ST3 Gal III sialyltransferase have been achieved in Aspergillus niger. At current levels of expression 300-450,000 liters of yeast fermentation would be required to produce

sufficient enzyme for sialylation of 1 kg of glycoprotein using the ST6Gal I sialyltransferase. In contrast, less than 10 liter fermentation of Aspergillus niger would be required for sialylation of 1 kg of glycoprotein using the ST3Gal III sialyltransferase. Thus, the fermentation capacity required to produce the ST3Gal III sialyltransferase for a large scale sialylation reaction would be 10-100 fold less than that required for producing the ST6Gal I; the cost of producing the sialyltransferase would be reduced proportionately.

PGPUB-DOCUMENT-NUMBER: 20040115168

PGPUB-FILING-TYPE: new

DOCUMENT-IDENTIFIER: US 20040115168 A1

TITLE: Interferon beta: remodeling and glycoconjugation of interferon beta

PUBLICATION-DATE: June 17, 2004

INVENTOR-INFORMATION:

| NAME | CITY | STATE | COUNTRY | RULE-47 |
|---|---|---|---|---|
| DeFrees, Shawn | North Wales | PA | US | |
| Zopf, David | Wayne | PA | US | |
| Bayer, Robert | San Diego | CA | US | |
| Bowe, Caryn | Doylestown | PA | US | |
| Hakes, David | Willow Grove | PA | US | |
| Chen, Xi | Lansdale | PA | US | |

APPL-NO: 10/ 410930

DATE FILED: April 9, 2003

RELATED-US-APPL-DATA:

child 10410930 A1 20030409

parent continuation-in-part-of 10360779 20030219 US PENDING

child 10410930 A1 20030409

parent continuation-in-part-of 10360770 20030106 US PENDING

child 10410930 A1 20030409

parent continuation-in-part-of 10287994 20021105 US PENDING

child 10287994 20021105 US

parent continuation-of PCT/US02/32263 20021009 US PENDING

non-provisional-of-provisional 60407527 20020828 US

non-provisional-of-provisional 60404249 20020816 US

non-provisional-of-provisional 60396594 20020717 US

non-provisional-of-provisional 60391777 20020625 US

non-provisional-of-provisional 60387292 20020607 US

non-provisional-of-provisional 60334301 20011128 US

non-provisional-of-provisional 60334233 20011128 US

non-provisional-of-provisional 60344692 20011019 US

non-provisional-of-provisional 60328523 20011010 US

US-CL-CURRENT: 424/85.6, 435/68.1 , 530/351

ABSTRACT:

The invention includes methods and compositions for remodeling a peptide molecule, including the addition or deletion of one or more glycosyl groups to a peptide, and/or the addition of a modifying group to a peptide.

CROSS REFERENCE TO RELATED APPLICATIONS

[0001] This application is a continuation-in-part of prior Application No. PCT/US02/32263, filed Oct. 9, 2002; Provisional Patent Application No. 60/448,381, filed Feb. 19, 2003 (converted to non-provisional application, same filing date, serial number not yet assigned); Provisional Patent Application No. 60/438,582, filed Jan. 6, 2003 (converted to non-provisional application, same filing date, serial number not yet assigned); Provisional Patent Application No. 60/407,527, filed Aug. 28, 2002; Provisional Patent Application No. 60/404,249, filed Aug. 16, 2002; Provisional Patent Application No. 60/396,594, filed Jul. 17, 2002; Provisional Patent Application No. 60/391,777, filed Jun. 25, 2002; Provisional Patent Application No. 60/387,292, filed Jun. 7, 2002; Provisional Patent Application No. 60/334,301, filed Nov. 28, 2001; Provisional Patent Application No. 60/334,233, filed Nov. 28, 2001; Provisional Patent Application No. 60/344,692, filed Oct. 19, 2001; and Provisional Patent Application No. 60/328,523, filed Oct. 10, 2001.

---------- KWIC ---------

Detail Description Paragraph - DETX (665):

[1034] Other sialyltransferases, including those listed in Table 8, are also useful in an economic and efficient large-scale process for sialylation of commercially important glycopeptides. As a simple test to find out the utility of these other enzymes, various amounts of each enzyme (1-100 mU/mg protein) are reacted with asialo-.alpha..sub.1 AGP (at 1-10 mg/ml) to compare the ability of the sialyltransferase of interest to sialylate glycopeptides relative to either bovine ST6Gal I, ST3Gal III or both sialyltransferases. Alternatively, other glycopeptides or glycopeptides, or N-linked oligosaccharides enzymatically released from the peptide backbone can be used in place of asialo-.alpha..sub.1 AGP for this evaluation. Sialyltransferases with the ability to sialylate N-linked oligosaccharides of glycopeptides more efficiently than ST6Gal I are useful in a practical large-scale process for peptide sialylation (as illustrated for ST3Gal III in this disclosure).

Detail Description Paragraph - DETX (1043):

[1411] This Example demonstrates that sialylation of recombinant glycoproteins using the ST3 Gal III sialyltransferase required much less enzyme than expected. For a one kilogram scale reaction, approximately 7,000 units of the ST3Gal III sialyltransferase would be needed, instead of 100,000-150,000 units that earlier studies indicated. Purification of these enzymes from natural sources is prohibitive, with yields of only 1-10 units for a large scale preparation after 1-2 months work. Assuming that both the ST6Gal I and ST3Gal III sialyltransferases are produced as recombinant sialyltransferases, with equal levels of expression of the two enzymes being achieved, a fermentation scale 14-21 times greater (or more) would be required for the ST6Gal I sialyltransferase relative to the ST3Gal III sialyltransferase. For the ST6Gal I sialyltransferase, expression levels of 0.3 U/l in yeast has been

reported (Borsig et al. (1995) Biochem. Biophys. Res. Commun. 210: 14-20). Expression levels of 1000 U/liter of the ST3 Gal III sialyltransferase have been achieved in Aspergillus niger. At current levels of expression 300-450,000 liters of yeast fermentation would be required to produce sufficient enzyme for sialylation of 1 kg of glycoprotein using the ST6Gal I sialyltransferase. In contrast, less than 10 liter fermentation of Aspergillus niger would be required for sialylation of 1 kg of glycoprotein using the ST3Gal III sialyltransferase. Thus, the fermentation capacity required to produce the ST3Gal III sialyltransferase for a large scale sialylation reaction would be 10-100 fold less than that required for producing the ST6Gal I; the cost of producing the sialyltransferase would be reduced proportionately.

PGPUB-DOCUMENT-NUMBER: 20040082026

PGPUB-FILING-TYPE: new

DOCUMENT-IDENTIFIER: US 20040082026 A1

TITLE: Interferon alpha: remodeling and glycoconjugation of interferon alpha

PUBLICATION-DATE: April 29, 2004

INVENTOR-INFORMATION:

| NAME | CITY | STATE | COUNTRY | RULE-47 |
|---|---|---|---|---|
| DeFrees, Shawn | North Wales | PA | US | |
| Zopf, David | Wayne | PA | US | |
| Bayer, Robert | San Diego | CA | US | |
| Bowe, Caryn | Doylestown | PA | US | |
| Hakes, David | Willow Grove | PA | US | |
| Chen, Xi | Lansdale | PA | US | |

APPL-NO: 10/ 411049

DATE FILED: April 9, 2003

RELATED-US-APPL-DATA:

child 10411049 A1 20030409

parent continuation-in-part-of 10360779 20030219 US PENDING

child 10411049 A1 20030409

parent continuation-in-part-of 10360770 20030106 US PENDING

child 10411049 A1 20030409

parent continuation-in-part-of 10287994 20021105 US PENDING

child 10287994 20021105 US

parent continuation-of PCT/US02/32263 20021009 US PENDING

non-provisional-of-provisional 60407527 20020828 US

non-provisional-of-provisional 60404249 20020816 US

non-provisional-of-provisional 60396594 20020717 US

non-provisional-of-provisional 60391777 20020625 US

non-provisional-of-provisional 60387292 20020607 US

non-provisional-of-provisional 60334301 20011128 US

non-provisional-of-provisional 60334233 20011128 US

non-provisional-of-provisional 60344692 20011019 US

non-provisional-of-provisional 60328523 20011010 US

US-CL-CURRENT: 435/68.1, 530/351

ABSTRACT:

The invention includes a multitude of methods and compositions for remodeling a peptide molecule, including the addition or deletion of one or more glycosyl groups to a peptide, and/or the addition of a modifying group to a peptide.

CROSS REFERENCE TO RELATED APPLICATIONS

[0001] This application is a continuation-in-part of prior Application No. PCT/US02/32263, filed Oct. 9, 2002; Provisional Patent Application No. 60/448,381, filed Feb. 19, 2003 (converted to non-provisional application, same filing date, serial number not yet assigned); Provisional Patent Application No. 60/438,582, filed Jan. 6, 2003 (converted to non-provisional application, same filing date, serial number not yet assigned); Provisional Patent Application No. 60/407,527, filed Aug. 28, 2002; Provisional Patent Application No. 60/404,249, filed Aug. 16, 2002; Provisional Patent Application No. 60/396,594, filed Jul. 17, 2002; Provisional Patent Application No. 60/391,777, filed Jun. 25, 2002; Provisional Patent Application No. 60/387,292, filed Jun. 7, 2002; Provisional Patent Application No. 60/334,301, filed Nov. 28, 2001; Provisional Patent Application No. 60/334,233, filed Nov. 28, 2001; Provisional Patent Application No. 60/344,692, filed Oct. 19, 2001; and Provisional Patent Application No. 60/328,523, filed Oct. 10, 2001.

---------- KWIC ---------

Detail Description Paragraph - DETX (671):
[1075] Other sialyltransferases, including those listed in Table 8, are also useful in an economic and efficient large-scale process for sialylation of commercially important glycopeptides. As a simple test to find out the utility of these other enzymes, various amounts of each enzyme (1-100 mU/mg protein) are reacted with asialo-.alpha..sub.1 AGP (at 1-10 mg/ml) to compare the ability of the sialyltransferase of interest to sialylate glycopeptides relative to either bovine ST6Gal I, ST3Gal III or both sialyltransferases. Alternatively, other glycopeptides or glycopeptides, or N-linked oligosaccharides enzymatically released from the peptide backbone can be used in place of asialo-.alpha..sub.1 AGP for this evaluation. Sialyltransferases with the ability to sialylate N-linked oligosaccharides of glycopeptides more efficiently than ST6Gal I are useful in a practical large-scale process for peptide sialylation (as illustrated for ST3Gal III in this disclosure).

Detail Description Paragraph - DETX (1049):
[1452] This Example demonstrates that sialylation of recombinant glycoproteins using the ST3 Gal III sialyltransferase required much less enzyme than expected. For a one kilogram scale reaction, approximately 7,000 units of the ST3Gal III sialyltransferase would be needed, instead of 100,000-150,000 units that earlier studies indicated. Purification of these enzymes from natural sources is prohibitive, with yields of only 1-10 units for a large scale preparation after 1-2 months work. Assuming that both the ST6Gal I and ST3Gal III sialyltransferases are produced as recombinant sialyltransferases, with equal levels of expression of the two enzymes being achieved, a fermentation scale 14-21 times greater (or more) would be required for the ST6Gal I sialyltransferase relative to the ST3Gal III sialyltransferase. For the ST6Gal I sialyltransferase, expression levels of 0.3 U/l in yeast has been

reported (Borsig et al. (1995) Biochem. Biophys. Res. Commun. 210: 14-20). Expression levels of 1000 U/liter of the ST3 Gal III sialyltransferase have been achieved in Aspergillus niger. At current levels of expression 300-450,000 liters of yeast fermentation would be required to produce sufficient enzyme for sialylation of 1 kg of glycoprotein using the ST6Gal I sialyltransferase. In contrast, less than 10 liter fermentation of Aspergillus niger would be required for sialylation of 1 kg of glycoprotein using the ST3Gal III sialyltransferase. Thus, the fermentation capacity required to produce the ST3Gal III **sialyltransferase for a large scale sialylation** reaction would be 10-100 fold less than that required for producing the ST6Gal I; the cost of producing the sialyltransferase would be reduced proportionately.

PGPUB-DOCUMENT-NUMBER: 20040077836

PGPUB-FILING-TYPE: new

DOCUMENT-IDENTIFIER: US 20040077836 A1

TITLE: Granulocyte colony stimulating factor: remodeling and glycoconjugation of G-CSF

PUBLICATION-DATE: April 22, 2004

INVENTOR-INFORMATION:

| NAME | CITY | STATE | COUNTRY | RULE-47 |
|---|---|---|---|---|
| DeFrees, Shawn | North Wales | PA | US | |
| Zopf, David | Wayne | PA | US | |
| Bayer, Robert | San Diego | CA | US | |
| Bowe, Caryn | Doylestown | PA | US | |
| Hakes, David | Willow Grove | PA | US | |
| Chen, Xi | Lansdale | PA | US | |

APPL-NO: 10/ 410962

DATE FILED: April 9, 2003

RELATED-US-APPL-DATA:

child 10410962 A1 20030409

parent continuation-in-part-of 10360779 20030219 US PENDING

child 10410962 A1 20030409

parent continuation-in-part-of 10360770 20030106 US PENDING

child 10360770 20030106 US

parent continuation-in-part-of 10287994 20021105 US PENDING

child 10287994 20021105 US

parent continuation-of PCT/US02/32263 20021009 US PENDING

non-provisional-of-provisional 60407527 20020828 US

non-provisional-of-provisional 60404249 20020816 US

non-provisional-of-provisional 60396594 20020717 US

non-provisional-of-provisional 60391777 20020625 US

non-provisional-of-provisional 60387292 20020607 US

non-provisional-of-provisional 60334301 20011128 US

non-provisional-of-provisional 60334233 20011128 US

non-provisional-of-provisional 60344692 20011019 US

non-provisional-of-provisional 60328523 20011010 US

US-CL-CURRENT: 530/351, 435/68.1

ABSTRACT:

The invention includes methods and compositions for remodeling a peptide molecule, including the addition or deletion of one or more glycosyl groups to a peptide, and/or the addition of a modifying group to a peptide.

CROSS REFERENCE TO RELATED APPLICATIONS

[0001] This application is a continuation-in-part of prior Application No. PCT/US02/32263, filed Oct. 9, 2002; Provisional Patent Application No. 60/448,381, filed Feb. 19, 2003 (converted to non-provisional application, same filing date, serial number not yet assigned); Provisional Patent Application No. 60/438,582, filed Jan. 6, 2003 (converted to non-provisional application, same filing date, serial number not yet assigned); Provisional Patent Application No. 60/407,527, filed Aug. 28, 2002; Provisional Patent Application No. 60/404,249, filed Aug. 16, 2002; Provisional Patent Application No. 60/396,594, filed Jul. 17, 2002; Provisional Patent Application No. 60/391,777, filed Jun. 25, 2002; Provisional Patent Application No. 60/387,292, filed Jun. 7, 2002; Provisional Patent Application No. 60/334,301, filed Nov. 28, 2001; Provisional Patent Application No. 60/334,233, filed Nov. 28, 2001; Provisional Patent Application No. 60/344,692, filed Oct. 19, 2001; and Provisional Patent Application No. 60/328,523, filed Oct. 10, 2001.

---------- KWIC ---------

Detail Description Paragraph - DETX (665):
[1001] Other sialyltransferases, including those listed in Table 8, are also useful in an economic and efficient large-scale process for sialylation of commercially important glycopeptides. As a simple test to find out the utility of these other enzymes, various amounts of each enzyme (1-100 mU/mg protein) are reacted with asialo-.alpha..sub.1 AGP (at 1-10 mg/ml) to compare the ability of the sialyltransferase of interest to sialylate glycopeptides relative to either bovine ST6Gal I, ST3Gal III or both sialyltransferases. Alternatively, other glycopeptides or glycopeptides, or N-linked oligosaccharides enzymatically released from the peptide backbone can be used in place of asialo-.alpha..sub.1 AGP for this evaluation. Sialyltransferases with the ability to sialylate N-linked oligosaccharides of glycopeptides more efficiently than ST6Gal I are useful in a practical large-scale process for peptide sialylation (as illustrated for ST3Gal III in this disclosure).

Detail Description Paragraph - DETX (1044):
[1379] This Example demonstrates that sialylation of recombinant glycoproteins using the ST3 Gal III sialyltransferase required much less enzyme than expected. For a one kilogram scale reaction, approximately 7,000 units of the ST3Gal III sialyltransferase would be needed, instead of 100,000-150,000 units that earlier studies indicated. Purification of these enzymes from natural sources is prohibitive, with yields of only 1-10 units for a large scale preparation after 1-2 months work. Assuming that both the ST6Gal I and ST3Gal III sialyltransferases are produced as recombinant sialyltransferases, with equal levels of expression of the two enzymes being achieved, a fermentation scale 14-21 times greater (or more) would be required for the ST6Gal I sialyltransferase relative to the ST3Gal III sialyltransferase. For the ST6Gal I sialyltransferase, expression levels of 0.3 U/i in yeast has been

reported (Borsig et al. (1995) Biochem. Biophys. Res. Commun. 210: 14-20). Expression levels of 1000 U/liter of the ST3 Gal III sialyltransferase have been achieved in Aspergillus niger. At current levels of expression 300-450,000 liters of yeast fermentation would be required to produce sufficient enzyme for sialylation of 1 kg of glycoprotein using the ST6Gal I sialyltransferase. In contrast, less than 10 liter fermentation of Aspergillus niger would be required for sialylation of 1 kg of glycoprotein using the ST3Gal III sialyltransferase. Thus, the fermentation capacity required to produce the ST3Gal III **sialyltransferase for a large scale sialylation** reaction would be 10-100 fold less than that required for producing the ST6Gal I; the cost of producing the sialyltransferase would be reduced proportionately.

PGPUB-DOCUMENT-NUMBER: 20040063911

PGPUB-FILING-TYPE: new

DOCUMENT-IDENTIFIER: US 20040063911 A1

TITLE: Protein remodeling methods and proteins/peptides produced by the methods

PUBLICATION-DATE: April 1, 2004

INVENTOR-INFORMATION:

| NAME | CITY | STATE | COUNTRY | RULE-47 |
|---|---|---|---|---|
| DeFrees, Shawn | North Wales | PA | US | |
| Zopf, David | Wayne | PA | US | |
| Bayer, Robert | San Diego | CA | US | |
| Hakes, David | Willow Grove | PA | US | |
| Chen, Xi | Lansdale | PA | US | |

APPL-NO: 10/ 411026

DATE FILED: April 9, 2003

RELATED-US-APPL-DATA:

child 10411026 A1 20030409

parent continuation-in-part-of 10360779 20030219 US PENDING

child 10360779 20030219 US

parent continuation-in-part-of 10360770 20030106 US PENDING

child 10360770 20030106 US

parent continuation-in-part-of 10287994 20021105 US PENDING

child 10287994 20021105 US

parent continuation-of PCT/US02/32263 20021009 US PENDING

non-provisional-of-provisional 60407527 20020828 US

non-provisional-of-provisional 60404249 20020816 US

non-provisional-of-provisional 60396594 20020717 US

non-provisional-of-provisional 60391777 20020625 US

non-provisional-of-provisional 60387292 20020607 US

non-provisional-of-provisional 60334301 20011128 US

non-provisional-of-provisional 60334233 20011128 US

non-provisional-of-provisional 60344692 20011019 US

non-provisional-of-provisional 60328523 20011010 US

US-CL-CURRENT: 530/351, 435/68.1 , 530/395

ABSTRACT:

The invention includes methods and compositions for remodeling a peptide molecule, including the addition or deletion of one or more glycosyl groups to a peptide, and/or the addition of a modifying group to a peptide.

CROSS REFERENCE TO RELATED APPLICATIONS

[0001] This application is a continuation-in-part of prior Application No. PCT/US02/32263, filed Oct. 9, 2002; Provisional Patent Application No. 60/448,381, filed Feb. 19, 2003 (converted to non-provisional application, same filing date, serial number not yet assigned); Provisional Patent Application No. 60/438,582, filed Jan. 6, 2003 (converted to non-provisional application, same filing date, serial number not yet assigned); Provisional Patent Application No. 60/407,527, filed Aug. 28, 2002; Provisional Patent Application No. 60/404,249, filed Aug. 16, 2002; Provisional Patent Application No. 60/396,594, filed Jul. 17, 2002; Provisional Patent Application No. 60/391,777, filed Jun. 25, 2002; Provisional Patent Application No. 60/387,292, filed Jun. 7, 2002; Provisional Patent Application No. 60/334,301, filed Nov. 28, 2001; Provisional Patent Application No. 60/334,233, filed Nov. 28, 2001; Provisional Patent Application No. 60/344,692, filed Oct. 19, 2001; and Provisional Patent Application No. 60/328,523, filed Oct. 10, 2001.

---------- KWIC ---------

Detail Description Paragraph - DETX (668):
[0950] Other sialyltransferases, including those listed in Table 8, are also useful in an economic and efficient large-scale process for sialylation of commercially important glycopeptides. As a simple test to find out the utility of these other enzymes, various amounts of each enzyme (1-100 mU/mg protein) are reacted with asialo-.alpha..sub.1 AGP (at 1-10 mg/ml) to compare the ability of the sialyltransferase of interest to sialylate glycopeptides relative to either bovine ST6Gal I, ST3Gal III or both sialyltransferases. Alternatively, other glycopeptides or glycopeptides, or N-linked oligosaccharides enzymatically released from the peptide backbone can be used in place of asialo-.alpha..sub.1 AGP for this evaluation. Sialyltransferases with the ability to sialylate N-linked oligosaccharides of glycopeptides more efficiently than ST6Gal I are useful in a practical large-scale process for peptide sialylation (as illustrated for ST3Gal III in this disclosure).

Detail Description Paragraph - DETX (1047):
[1328] This Example demonstrates that sialylation of recombinant glycoproteins using the ST3 Gal III sialyltransferase required much less enzyme than expected. For a one kilogram scale reaction, approximately 7,000 units of the ST3Gal III sialyltransferase would be needed, instead of 100,000-150,000 units that earlier studies indicated. Purification of these enzymes from natural sources is prohibitive, with yields of only 1-10 units for a large scale preparation after 1-2 months work. Assuming that both the ST6Gal I and ST3Gal III sialyltransferases are produced as recombinant sialyltransferases, with equal levels of expression of the two enzymes being achieved, a fermentation scale 14-21 times greater (or more) would be required for the ST6Gal I sialyltransferase relative to the ST3Gal III sialyltransferase. For the ST6Gal I sialyltransferase, expression levels of 0.3 U/l in yeast has been reported (Borsig et al. (1995) Biochem. Biophys. Res. Commun. 210: 14-20).

Expression levels of 1000 U/liter of the ST3 Gal III sialyltransferase have been achieved in Aspergillus niger. At current levels of expression 300-450,000 liters of yeast fermentation would be required to produce sufficient enzyme for sialylation of 1 kg of glycoprotein using the ST6Gal I sialyltransferase. In contrast, less than 10 liter fermentation of Aspergillus niger would be required for sialylation of 1 kg of glycoprotein using the ST3Gal III sialyltransferase. Thus, the fermentation capacity required to produce the ST3Gal III sialyltransferase for a large scale sialylation reaction would be 10-100 fold less than that required for producing the ST6Gal I; the cost of producing the sialyltransferase would be reduced proportionately.

PGPUB-DOCUMENT-NUMBER: 20040043446

PGPUB-FILING-TYPE: new

DOCUMENT-IDENTIFIER: US 20040043446 A1

TITLE: Alpha galalctosidase a: remodeling and glycoconjugation of alpha galactosidase A

PUBLICATION-DATE: March 4, 2004

INVENTOR-INFORMATION:

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APPL-NO: 10/ 411037

DATE FILED: April 9, 2003

RELATED-US-APPL-DATA:

child 10411037 A1 20030409

parent continuation-in-part-of PCT/US02/32263 20021009 US PENDING

non-provisional-of-provisional 60407527 20020828 US

non-provisional-of-provisional 60404249 20020816 US

non-provisional-of-provisional 60396594 20020717 US

non-provisional-of-provisional 60391777 20020625 US

non-provisional-of-provisional 60387292 20020607 US

US-CL-CURRENT: 435/68.1, 435/193 , 435/208

ABSTRACT:

The invention includes methods and compositions for remodeling a peptide molecule, including the addition or deletion of one or more glycosyl groups to a peptide, and/or the addition of a modifying group to a peptide.

CROSS REFERENCE TO RELATED APPLICATIONS

[0001] This application is a continuation-in-part of prior Application No. PCT/US02/32263, filed Oct. 9, 2002; Provisional Patent Application No. 60/448,381, filed Feb. 19, 2003 (converted to non-provisional application, same filing date, serial number not yet assigned); Provisional Patent Application No. 60/438,582, filed Jan. 6, 2003 (converted to non-provisional application, same filing date, serial number not yet assigned); Provisional Patent Application No. 60/407,527, filed Aug. 28, 2002; Provisional Patent Application No. 60/404,249, filed Aug. 16, 2002; Provisional Patent

Application No. 60/396,594, filed Jul. 17, 2002; Provisional Patent Application No. 60/391,777, filed Jun. 25, 2002; Provisional Patent Application No. 60/387,292, filed Jun. 7, 2002; Provisional Patent Application No. 60/334,301, filed Nov. 28, 2001; Provisional Patent Application No. 60/334,233, filed Nov. 28, 2001; Provisional Patent Application No. 60/344,692, filed Oct. 19, 2001; and Provisional Patent Application No. 60/328,523, filed Oct. 10, 2001.

---------- KWIC ---------

Detail Description Paragraph - DETX (660):

[1025] Other sialyltransferases, including those listed in Table 8, are also useful in an economic and efficient large-scale process for sialylation of commercially important glycopeptides. As a simple test to find out the utility of these other enzymes, various amounts of each enzyme (1-100 mU/mg protein) are reacted with asialo-.alpha..sub.1 AGP (at 1-10 mg/ml) to compare the ability of the sialyltransferase of interest to sialylate glycopeptides relative to either bovine ST6Gal I, ST3Gal III or both sialyltransferases. Alternatively, other glycopeptides or glycopeptides, or N-linked oligosaccharides enzymatically released from the peptide backbone can be used in place of asialo-.alpha..sub.1 AGP for this evaluation. Sialyltransferases with the ability to sialylate N-linked oligosaccharides of glycopeptides more efficiently than ST6Gal I are useful in a practical large-scale process for peptide sialylation (as illustrated for ST3Gal III in this disclosure).

Detail Description Paragraph - DETX (1036):

[1400] This Example demonstrates that sialylation of recombinant glycoproteins using the ST3 Gal III sialyltransferase required much less enzyme than expected. For a one kilogram scale reaction, approximately 7,000 units of the ST3Gal III sialyltransferase would be needed, instead of 100,000-150,000 units that earlier studies indicated. Purification of these enzymes from natural sources is prohibitive, with yields of only 1-10 units for a large scale preparation after 1-2 months work. Assuming that both the ST6Gal I and ST3Gal III sialyltransferases are produced as recombinant sialyltransferases, with equal levels of expression of the two enzymes being achieved, a fermentation scale 14-21 times greater (or more) would be required for the ST6Gal I sialyltransferase relative to the ST3Gal III sialyltransferase. For the ST6Gal I sialyltransferase, expression levels of 0.3 U/l in yeast has been reported (Borsig et al. (1995) Biochem. Biophys. Res. Commun. 210: 14-20). Expression levels of 1000 U/liter of the ST3 Gal 1H sialyltransferase have been achieved in Aspergillus niger. At current levels of expression 300-450,000 liters of yeast fermentation would be required to produce sufficient enzyme for sialylation of 1 kg of glycoprotein using the ST6Gal I sialyltransferase. In contrast, less than 10 liter fermentation of Aspergillus niger would be required for sialylation of 1 kg of glycoprotein using the ST3Gal III sialyltransferase. Thus, the fermentation capacity required to produce the ST3Gal III sialyltransferase for a large scale sialylation reaction would be 10-100 fold less than that required for producing the ST6Gal I; the cost of producing the sialyltransferase would be reduced proportionately.

PGPUB-DOCUMENT-NUMBER: 20040002138

PGPUB-FILING-TYPE: new

DOCUMENT-IDENTIFIER: US 20040002138 A1

TITLE: Alpha,2,8-sialyltransferase

PUBLICATION-DATE: January 1, 2004

INVENTOR-INFORMATION:

| NAME | CITY | STATE | COUNTRY | RULE-47 |
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APPL-NO: 10/ 430325

DATE FILED: May 7, 2003

RELATED-US-APPL-DATA:

child 10430325 A1 20030507

parent division-of 08361304 19941129 US GRANTED

parent-patent 6596523 US

child 08361304 19941129 US

parent continuation-in-part-of PCT/JP94/00495 19940328 US UNKNOWN

FOREIGN-APPL-PRIORITY-DATA:

| COUNTRY | APPL-NO | DOC-ID | APPL-DATE |
|---|---|---|---|
| JP | HEI-5-69988 | 1993JP-HEI-5-69988 | March 29, 1993 |

US-CL-CURRENT: 435/69.1, 435/193 , 435/320.1 , 435/325 , 435/85 , 536/23.2 , 536/53

ABSTRACT:

The invention provides a novel .alpha.-2,8-sialyltransferase expressed by a gene cloned from animal cells, a cDNA coding for the .alpha.-2,8-sialyltransferase, a method of detecting, or suppressing the production of .alpha.-2,8-sialyltransferase by using the cDNA, a recombinant vector containing the DNA as an insert and cells harboring the recombinant vector as well as methods of preparing same. The .alpha.-2,8-sialyltransferase of the invention is useful, for example, in the production of carbohydrate chains having a useful physiological activity, for example the ganglioside GD3, and modifications thereof.

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Summary of Invention Paragraph - BSTX (22):

[0020] As far as sialyltransferase is concerned, a gene for an enzyme having .beta.-galactoside .alpha.-2,6-sialyltransferase activity has been isolated and

the base sequence thereof has been reported [Weinstein et al.: Journal of Biological Chemistry, 262, 17735 (1987)]. As regards an enzyme having .beta.-galactoside .alpha.-2,3-sialyltransferase activity, cloning of a gene coding for an enzyme catalyzing the addition of sialic acid to galactose in an O-glycoside bond type carbohydrate chain (carbohydrate chain added to a serine or threonine residue) of glycoproteins has been reported by Gillespie et al. but the base sequence of said gene has not been reported [Gillespie et al.: Glycoconjugate Journal, 7, 469 (1990)]. Weinstein et al. reported a method of purifying an enzyme having .beta.-galactoside .alpha.-2,3-sialyltransferase activity from rat liver [Weinstein et al.: Journal of Biological Chemistry, 257, 13835 (1982)]. This method, however, provides the desired enzyme only in very small amounts. This rat liver .beta.-galactoside .alpha.-2,3-sialyltransferase gene has been cloned by Wen et al. [Wen et al.: Journal of Biological Chemistry, 267, 21011 (1992)]. There has been no report, however, of the cloning of a gene for human galactoside .alpha.-2,8-sialyltransferase. Large scale preparation of a sialyltransferase species having .alpha.-2,8-sialyltransferase activity or cloning of a gene for encoding a product having sialyltransferase activity has not been reported as yet. Therefore, no means is currently available for large scale preparation of a sialyltransferase having .alpha.-2,8-sialyltransferase activity, in particular human galactoside .alpha.-2,8-sialyltransferase. Methods of detecting or suppressing expression of the enzyme have also not been established.

US-PAT-NO: 6709834

DOCUMENT-IDENTIFIER: US 6709834 B2

TITLE: Lipopolysaccharide .alpha.-2,3 sialytransferase of campylobacter jejuni and its uses

DATE-ISSUED: March 23, 2004

INVENTOR-INFORMATION:

| NAME | CITY | STATE | ZIP CODE | COUNTRY |
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APPL-NO: 10/ 058636

DATE FILED: January 29, 2002

PARENT-CASE:

CROSS-REFERENCE TO RELATED APPLICATION

This application is a divisional of and claims benefit of U.S. application Ser. No. 09/272,960, filed Mar. 18, 1999, which claims the benefit of U.S. Provisional Application No. 60/078,891, filed Mar. 20, 1998 now abandoned, the disclosures of both of which are incorporated herein by reference, in their entirety for all purposes.

US-CL-CURRENT: 435/15, 435/183 , 435/193 , 435/220 , 435/252.3 , 435/320.1 , 435/4 , 435/41 , 435/6 , 435/7.2 , 435/85 , 435/97 , 530/350

ABSTRACT:

The structure and specificity of a recombinant .alpha.2,3-sialyltransferase from Campylobacter spp., is disclosed. Also provided are methods for using the .alpha.2,3-sialyltransferase in the production of desired carbohydrate structures and nucleic acids that encode the sialyltransferase.

15 Claims, 3 Drawing figures

Exemplary Claim Number: 1

Number of Drawing Sheets: 4

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Other Reference Publication - OREF (3):
Williams et al., "Large-scale expression of recombinant sialyltransferases and comparison of their kinetic properties with native enzymes," Glycoconjugate J. 12: 755-761.

US-PAT-NO: 6706497

DOCUMENT-IDENTIFIER: US 6706497 B2

TITLE: Methods for producing sialyloligosaccharides in a dairy source

DATE-ISSUED: March 16, 2004

INVENTOR-INFORMATION:

| NAME | CITY | STATE | ZIP CODE | COUNTRY |
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| Barker; William A. | West Chester | PA | N/A | N/A |
| Hakes; David J. | Willow Grove | PA | N/A | N/A |
| Zopf; David A. | Strafford | PA | N/A | N/A |


APPL-NO: 09/ 955909

DATE FILED: September 18, 2001

PARENT-CASE:

This is a continuation of application Ser. No. 08/911,393, filed Aug. 14, 1997 now U.S. Pat. No. 6,323,008.

US-CL-CURRENT: 435/84, 435/101 , 435/274 , 435/99 , 536/124 , 536/127

ABSTRACT:

The present invention provides methods for producing sialyloligosaccharides in situ in dairy sources and cheese processing waste streams, prior to, during, or after processing of the dairy source during the cheese manufacturing process. The methods of the present invention use the catalytic activity of .alpha.(2-3) trans-sialidases to exploit the high concentrations of lactose and .alpha.(2-3) sialosides which naturally occur in dairy sources and cheese processing waste streams to drive the enzymatic synthesis of .alpha.(2-3) sialyllactose. .alpha.(2-3) sialyloligosaccharides produced according to these methods are additionally encompassed by the present invention. The invention also provides for recovery of the sialyloligosaccharides produced by these methods. The invention further provides a method for producing .alpha.(2-3) sialyllactose. The invention additionally provides a method of enriching for .alpha.(2-3) sialyllactose in milk using transgenic mammals that express an .alpha.(2-3) trans-sialidase transgene. The invention also provides for recovery of the sialyllactose contained in the milk produced by this transgenic mammal either before or after processing of the milk. Transgenic mammals containing an .alpha.(2-3) trans-sialidase encoding sequence operably linked to a regulatory sequence of a gene expressed in mammary tissue are also provided by the invention.

36 Claims, 10 Drawing figures

Exemplary Claim Number: 1

Number of Drawing Sheets: 10

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Brief Summary Text - BSTX (27):

Numerous foreign proteins have successfully been transgenically expressed in the milk of livestock. Most of this work has focused on the expression of proteins which are foreign to the mammary gland. Colman, A., 1996, Am. J. Clin. Nutr. 63:639S-645S. To date, milk specific expression of transgenic livestock has been achieved through operably linking regulatory sequences of milk-specific protein genes to the target protein-encoding gene sequence, microinjecting these genetic constructs into the pronuclei of fertilized embryos, and implanting the embryos into recipient females. See e.g. Wright et al., 1991, Biotechnology (NY) 9:830-834; Carver et al., 1993, Biotechnology (NY) 11:1263-1270; Paterson et al., 1994, Appl. Microbiol. Biotechnol. 40:691-698. Proteins that have been successfully expressed in the milk of transgenic animals, include: .alpha.1-antitrypsin (Wright et al., 1991, Biotechnology (NY) 9:830-834; Carver et al., 1993, Biotechnology (NY) 11:1263-1270); Factor IX (Clark et al., 1989, Biotechnology (NY) 7:487-492); protein C (Velander et al., 1992, Proc. Natl. Acad. Sci. USA, 89:12003-12007); tissue plasminogen activator (Ebert et al., 1991, Biotechology (NY) 9:835-838); and fibrinogen. While most of these transgenes express proteins that supplement the composition of milk, very few, if any of the expressed proteins interact directly with the components of milk to alter the natural milk composition. There is a need for methods providing for the large scale production of .alpha.(2-3) sialyloligosaccharides, such as .alpha.(2-3) sialyllactose, which have commercial and/or therapeutic valve.

Brief Summary Text - BSTX (29):

The present invention greatly advances the field of commercial production of sialyloligosaccharides by providing methods for producing sialyloligosaccharides in situ in dairy sources and cheese processing waste streams. The methods of the invention have particular applications in producing .alpha.(2-3) sialyllactose in a dairy source prior to, during, or after processing of the dairy source during the cheese manufacturing process, thereby greatly increasing the recoverable yield of .alpha.(2-3) sialyllactose from the dairy source.

US-PAT-NO: 6689604

DOCUMENT-IDENTIFIER: US 6689604 B1

TITLE: Lipopolysaccharide .alpha.-2,3 sialyltransferase of Campylobacter jejuni and its uses

DATE-ISSUED: February 10, 2004


INVENTOR-INFORMATION:

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| Wakarchuk; Warren W. | Gloucester | N/A | N/A | CA |



APPL-NO: 09/ 272960

DATE FILED: March 18, 1999


PARENT-CASE:

CROSS-REFERENCE TO RELATED APPLICATION

This application claims benefit of U.S. Provisional Application No. 60/078,891, filed Mar. 20, 1998, which application is incorporated herein by reference for all purposes.


US-CL-CURRENT: 435/320.1, 435/252.3 , 435/252.33 , 435/346 , 435/6 , 435/68.1 , 435/69.1 , 435/69.3 , 435/70.2 , 435/71.1 , 435/71.2 , 435/74 , 435/822 , 514/54 , 536/23.1 , 536/23.2 , 536/24.3



ABSTRACT:

The structure and specificity of a recombinant .alpha.2,3-sialyltransferase from Campylobacter spp., is disclosed. Also provided are methods for using the .alpha.2,3-sialyltransferase in the production of desired carbohydrate structures and nucleic acids that encode the sialyltransferase.


30 Claims, 3 Drawing figures

Exemplary Claim Number: 1

Number of Drawing Sheets: 4

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Other Reference Publication - OREF (12):

Williams et al., "Large-scale expression of recombinant sialyltransferases and comparison of their kinetic properties with native enzymes," Glycoconjugate J. 12: 755-761.